FORENSIC BIOLOGY SECTION

DATABASING TEST METHODS

FORWARD

The Laboratory Division of the Indiana State Police (ISP) maintains the Combined DNA Index System (CODIS) or DNA Database. The Laboratory organizes and funds the collection of legislatively mandated samples from convicted offenders. The analysts of the Biology Section shall have a minimum of a baccalaureate or an advanced degree in a natural science or a closely related field. DNA analysts shall have successfully completed college course work covering the subject areas of genetics, biochemistry, molecular biology and statistics. All analysts undergo an intensive formalized training program dealing with forensic techniques and instrumentation. Completion of the Training Program is required before analysis of databasing samples is performed. Additionally, all analysts participate in proficiency testing utilizing open trials, blind trials and/or re-examination techniques. The accuracy and specificity of test results are ensured by running known controls with each set of tests.

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1. LDIS (Local DNA Index System) Methods:

1.1 Scope:

This test method is designed for the guidance of laboratory personnel (at all four laboratories) who interact with the Combined DNA Index System (CODIS). The scope of this interaction may include but is not limited to submitting profiles from casework samples for entry into the database, performing CODIS Administrator functions, or being involved in hit confirmations and reporting. This test method may be expanded or altered as techniques, software and/or new legal authorities are found applicable.

1.1.1 Indiana Criminal Code 10-13-6 (Appendix 1) authorizes the Superintendent of the Indiana State Police (ISP) to establish and maintain the Indiana DNA Database. The purpose of this database is to assist federal, state and local criminal justice and law enforcement agencies in the putative identification, detection, or exclusion of individuals who are subjects of an investigation or prosecution of a crime in which biological evidence is recovered from a crime scene.

1.1.2 The Indiana DNA Database Administrative Rules (Appendix 2) assign the Commander of the Indiana State Police Laboratory (ISPL) the responsibility for the administration of the Indiana DNA Database subject to the authority and approval of the Superintendent.

1.1.3 In addition to these procedures, all current National DNA Index System (NDIS) Procedures, available on the Criminal Justice Information Services Division Wide Area Network (CJIS-WAN), shall be followed.

1.2 Precautions/Limitations:

1.2.1 Casework profiles submitted for entry into CODIS should be evaluated carefully for eligibility in accordance with National DNA Index System (NDIS) Operational Procedure "DNA Records Accepted at NDIS" and the flowchart "A Guide to Determining What is Allowable in the Forensic Index at NDIS," both available on the CJIS-WAN. Indiana Criminal Code 10-13-6 does not authorize databasing of additional categories of samples.

1.3 Related Information:

1.3.1 Indiana Criminal Code 10-13-6
1.3.2 NDIS Operational Procedures

1.3.3 Worksheet Manual

1.4 Instruments: Designated computer terminals and software

1.5 Reagents/Materials: None

1.6 Hazards/Safety: None

1.7 Reference Materials/Controls/Calibration Checks:

1.7.1 DNA profiles shall be developed in compliance with the DNA Identification Act of 1994 and the FBI Approved Quality Assurance Standards for Forensic DNA Testing Laboratories.

1.8 Procedures/Instructions:

1.8.1 CODIS Entry

1.8.1.1 Profiles for entry shall be recorded on a CODIS Information Worksheet and technically reviewed including accuracy of allele calls and specimen category.

1.8.1.2 Categories of DNA data eligible at LDIS are Forensic Unknown, Forensic Mixture, Forensic Partial, Missing Person, Deduced Missing Person, Relative of Missing Person and Unidentified Person.

1.8.1.2.1 Profiles entered from the CODIS Information Worksheet shall be entered as Forensic Unknown if the profile being entering is believed to be from a single source.

1.8.1.2.2 Profiles shall be entered as Forensic Mixture if the data being entered is believed to be a mixture of more than one individual. The match estimator shall be used to ensure the mixture satisfies a statistical threshold for rarity of approximately 1 in the size of NDIS.

1.8.1.2.3 Other specimen categories shall be used as appropriate according to their definitions in NDIS Operational Procedure "NDIS DNA Records" and indicated on the CODIS Information Worksheet.

1.8.1.3 The CODIS State or Local Administrator or other designated personnel shall evaluate and specify on the CODIS Information Worksheet which profile(s) or alleles shall be entered into CODIS.

1.8.1.4 All data specified for DNA database entry on the CODIS Information Worksheet shall be entered into CODIS. Double entry is required, but may be conducted by the same individual.

1.8.1.5 A Specimen Detail Report shall be printed for each profile entered from the CODIS Information Worksheet and placed with the case notes.

1.8.1.6 The Specimen Detail Report shall be reviewed to ensure entry of the correct profiles, accuracy of alleles and correct specimen category during review of the report. This shall be done by the Technical Reviewer.

1.8.1.7 CODIS input shall include all loci with conclusive types and shall not be limited to the core 13 STR loci.

1.8.1.8 All suitable data shall be marked for transfer to the State DNA Index System (SDIS) at the time of entry.

1.8.1.9 A CODIS administrator shall upload new or changed data on a routine basis.

1.8.1.10 If the ISPL has a current contract and/or is in compliance with Standard 17 of the Quality Assurance Audit for Forensic DNA and Convicted Offender DNA Databasing Laboratories concerning the private laboratory used, the contributing agency may request the profile(s) developed in the case be entered into CODIS.

1.8.1.10.1 The complete case record must be provided to the ISPL for technical review.

1.8.1.10.2 Data must be in a platform and test kit which ISP personnel are qualified and proficiency tested to review.

1.8.1.10.3 Prior to the start of analysis by the private laboratory, documented approval of all technical specifications and acceptance of ownership of the data must be obtained from the ISPL Technical Leader.

1.8.1.10.4 After technical review, qualifying profiles shall be entered into CODIS in the same manner as cases analyzed by ISP.

1.8.1.11 If the ISPL does not have a contract or current compliance with Standard 17 concerning the private laboratory and/or prior approval from the ISPL Technical Leader, profiles developed by that laboratory at the request of the contributing agency shall not be entered into CODIS.

1.8.2 Hit Confirmation and Reporting

1.8.2.1 Evaluation of Matches

1.8.2.1.1 An effort shall be made to resolve all matches within 30 business days, except in circumstances beyond the control of ISPL personnel.

1.8.2.1.2 High Stringency matches that provide previously unknown information to an unsolved case (as determined by the case record and/or contact with the investigator) shall be confirmed.

1.8.2.1.3 Moderate Stringency matches shall be reviewed by an individual with mixture interpretation training/experience. The case analyst may also be consulted to determine if the match should be confirmed or given a disposition of No Match.

1.8.2.1.4 The laboratory with ownership of an unsolved casework profile involved in a match has primary responsibility to request any needed confirmation of that match.

1.8.2.2 Offender to Case Hits

1.8.2.2.1 State DNA Index System (SDIS) Hits

1.8.2.2.1.1 Confirmation shall be requested from the SDIS laboratory.

1.8.2.2.1.2 The case profile shall be verified by reviewing the original DNA analysis data. This shall be recorded on the Hit Confirmation Checklist.

1.8.2.2.1.3 The match shall be verified by a qualified analyst. This shall be recorded on the Hit Confirmation Checklist.

1.8.2.2.1.4 The Hit Confirmation Checklist shall be returned to the SDIS laboratory.

1.8.2.2.1.5 The disposition and Source ID fields shall be updated.

1.8.2.2.2 National DNA Index System (NDIS) Hits

1.8.2.2.2.1 Confirmation shall be requested from the NDIS laboratory.

1.8.2.2.2.2 The SDIS laboratory shall be notified for hit counting purposes.

1.8.2.2.2.3 The case profile shall be verified by reviewing the original DNA analysis data. This shall be recorded on the Hit Confirmation Checklist.

1.8.2.2.2.4 The match shall be verified by a qualified analyst. This shall be recorded on the Hit Confirmation Checklist.

1.8.2.2.2.5 When all verifications are complete, the Hit Confirmed line on the Hit Confirmation Checklist shall be signed by the CODIS Administrator.

1.8.2.2.2.6 A Laboratory Information Management System (LIMS) report shall be generated stating the match results. No statistics shall be included in the hit report. Hit reports shall follow the CODIS Hit Report Wording as in 2.11.

1.8.2.2.2.7 The disposition and Source ID fields shall be updated.

1.8.2.3 Case to Case Hits

1.8.2.3.1 When at least one case is unsolved, confirmation shall be requested from the other laboratory involved in the match, if the cases are from different laboratories.

1.8.2.3.2 The SDIS laboratory shall be notified for hit counting purposes.

1.8.2.3.3 Each case profile shall be verified by reviewing the original DNA analysis data. This shall be recorded on the Hit Confirmation Checklist.

1.8.2.3.4 The match shall be verified by a qualified analyst. This shall be recorded on the Hit Confirmation Checklist.

1.8.2.3.5 When all verifications are complete, the Hit Confirmed line on the Hit Confirmation Checklist shall be signed by a CODIS Administrator at one of the laboratories involved in the match.

1.8.2.3.6 The cases shall be related in LIMS and a LIMS report shall be generated for each matching case profile from an ISPL case. Hit reports shall follow the CODIS Hit Report Wording as in 2.11.

1.8.2.3.7 If the match involves a laboratory from another state, a match response containing information similar to that which would be included in the LIMS report shall be forwarded to that laboratory.

1.8.2.3.7.1 This information shall be administratively reviewed for accuracy. This review shall be documented by the reviewer's initials on the page.

1.8.2.3.8 The disposition and Source ID fields, if appropriate, shall be updated.

1.8.2.4 In instances where it is determined that the offender is not incarcerated and there are unusual public safety concerns, the investigating agency may be given verbal notification of the match with the reminder that the hit confirmation is in process. The hit report shall not be issued until the match is confirmed.

1.8.2.5 The LIMS case report documenting the match may be written by the CODIS Unit at the Indianapolis Regional Laboratory for any ISPL case.

1.8.3 Systems Operations

1.8.3.1 Access to CODIS shall be limited to laboratory personnel designated by the CODIS Unit Supervisor.

1.8.3.2 Access by NDIS or NDIS contract personnel may be allowed for the purpose of diagnosing, repairing and servicing CODIS software. Any such access shall be documented on the Indiana State Police CODIS Remote Access Log.

1.8.3.3 All users shall log off or lock the CODIS computers after each session. Terminals shall lock after 10 minutes of non-use.

1.8.3.4 Backups of all files on the CODIS computers shall be performed at minimum once per week.

1.8.3.4.1 Backups may be documented by either automatic scheduling within backup software or a written log if done manually.

1.8.3.4.2 The backup media shall be stored in a secure location.

1.8.3.4.3 Backups shall be stored in a separate building in a secure area once per month.

1.8.3.4.4 The movement of backup media shall be recorded on the CODIS Backup Log.

1.9 Records:

1.9.1 The appropriate worksheets as contained in the Biology Section Worksheet Manual shall be used to record all procedures.

1.9.2 Documents relating to hit confirmations shall be maintained in the Hit File maintained by the CODIS Unit at the Indianapolis Regional Laboratory and/or in LIMS.

1.9.3 User documentation and backup logs shall be maintained by the CODIS Administrators at each laboratory.

1.10 Interpretation of Results: All Candidate Matches shall be reviewed and evaluated by a DNA analyst currently or previously qualified in the technology being reviewed. Matches shall be given a disposition in accordance with the disposition definitions provided in current NDIS Operational Procedures.

1.11 Report Writing: See section 2.11

1.12 References: None

2 SDIS (State DNA Index System) Methods

2.1 Scope:

This test method is designed for the guidance of CODIS Unit personnel who interact with the Combined DNA Index System (CODIS). The scope of this interaction may include but is not limited to entering profiles from casework or offenders, accessioning and preparing offender samples, performing CODIS Administrator functions, or being involved in hit confirmations and reporting. This test method may be expanded or altered as techniques, software and/or new legal authorities are found applicable.

2.1.1 Indiana Criminal Code 10-13-6 (Appendix 1) authorizes the Superintendent of the Indiana State Police (ISP) to establish and maintain the Indiana DNA Database. The purpose of this database is to assist federal, state and local criminal justice and law enforcement agencies in the putative identification, detection, or exclusion of individuals who are subjects of an investigation or prosecution of a crime in which biological evidence is recovered from a crime scene.

2.1.2 The Indiana DNA Database Administrative Rules (Appendix 2) assign the Commander of the Indiana State Police Laboratory (ISPL) the responsibility for the administration of the Indiana DNA Database subject to the authority and approval of the Superintendent.

2.1.3 In addition to these procedures, all current National DNA Index System (NDIS) Procedures, available on the Criminal Justice Information Services Division Wide Area Network (CJIS-WAN), shall be followed.

2.2 Precautions/Limitations:

2.2.1 Profiles submitted for entry into CODIS should be evaluated carefully for eligibility in accordance with National DNA Index System (NDIS) Operational Procedure "DNA Records Accepted at NDIS" and the flowchart "A Guide to Determining What is Allowable in the Forensic Index at NDIS," both available on the CJIS-WAN. Indiana Criminal Code 10-13-6 does not authorize databasing of additional categories of samples.

2.2.2 Convicted Offender samples collected for Combined DNA Index System (CODIS) shall be treated as reference materials and not considered evidence.

2.3 Related Information:

2.3.1 Indiana Criminal Code 10-13-6
2.3.2 Indiana DNA Database Administrative Rules
2.3.3 NDIS Operational Procedures
2.3.4 Worksheet Manual

2.4 Instruments: Designated computer terminals and software

2.5 Reagents/Materials: None

2.6 Hazards/Safety:

2.6.1 Universal Precautions shall be used whenever biological materials are being handled.

2.6.2 Biological waste shall be disposed of in the appropriate waste receptacle.

2.7 Reference Materials/Controls/Calibration Checks:

2.7.1 DNA profiles shall be developed in compliance with the DNA Identification Act of 1994, the FBI Approved Quality Assurance Standards for Forensic DNA Testing Laboratories and the FBI Approved Quality Assurance Standards for DNA Databasing Laboratories.

2.7.2 Convicted Offenders who are collected and analyzed more than once shall be compared to ensure their DNA profiles and identities match. An effort shall be made to resolve any discrepancies.

2.8 Procedures/Instructions:

2.8.1 Collection and Delivery of Convicted Offender Samples

2.8.1.1 DNA samples shall be collected in a medically approved manner by a physician, registered nurse, licensed vocational nurse, licensed clinical technologist, or other person trained to properly collect DNA samples.

2.8.1.2 The Department of Correction (DOC) Reception and Diagnostic Center and the DOC Indiana Women's Prison shall collect a DNA sample from all convicted felons entering the DOC. The county sheriff or their

designee shall collect a sample from all felons not entering DOC. A private vendor may be used.

2.8.1.3 At the time of sample collection the collecting agency shall complete an Indiana Offender DNA Database Sample Information Sheet, State Form 47808 or another approved form. The name and date of birth, DOC inmate number, social security number or other identifying number shall be placed on the Sample Information Sheet. The right thumbprint of the offender shall be placed on the Sample Information Sheet. If no right thumb is present, the print of another finger shall be placed on the Sample Information Sheet with notation indicating the finger used.

2.8.1.4 The person who collected the sample shall seal offender samples in appropriate containers. Samples should be stored in a secure location prior to delivery to the ISPL.

2.8.1.5 Prior to delivery, the offender name and/or identifying number for all samples being delivered that day shall be listed on the Indiana Offender DNA Data Base Sample Delivery Receipt, State Form 47809 or another approved form. The person who took the samples shall prepare and sign the receipt. The collecting agency may retain a copy of the Sample Delivery Receipt. If a private vendor is used, they shall not retain a copy.

2.8.1.6 DOC samples listed on the receipt shall be placed in a locking container along with the top two copies of the receipt. The DOC collecting employee shall lock the container for hand transport to the ISPL. Samples collected by a private vendor shall be delivered to the ISPL via a carrier that uses a package tracking system.

2.8.1.7 At the ISPL the container shall be unlocked or opened and the contents removed. Laboratory staff shall count the samples to ensure that the number of samples matches the number of samples listed on the receipt.

2.8.1.7.1 If the number of samples in the container and the number of samples listed on the receipt do not match, a notation shall be made on the receipt. The discrepancy shall be resolved by contacting the collecting agency.

2.8.1.8 The DOC correctional officer delivering the samples and the ISPL personnel receiving the samples shall sign the receipt. The top copy of the receipt shall be retained by the ISPL. The second copy shall be returned to the container and locked for return to the DOC.

2.8.1.9 Samples not delivered in person shall be sent to the ISPL via a carrier that uses a package tracking system.

2.8.2 Handling and Storage of Convicted Offender Samples

2.8.2.1 Samples not immediately processed shall be stored in a secure location.

2.8.2.2 The offender name and/or identifying number of the sample may be compared to the ISP Convicted Offender sample tracking database of previously sampled offenders.

2.8.2.2.1 Samples from offenders not previously in the tracking database shall be assigned a new Indiana Offender Database (INODB) number and added to the tracking database. The INODB number is a unique, non-identifying number assigned by the ISPL. INODB numbers start at 000001 and are assigned consecutively.

2.8.2.2.2 Samples from offenders previously in the tracking database may be saved for potential use as quality assurance samples.

2.8.2.2.2.1 If retained as a quality assurance sample, the sample shall be given a new INODB number and added to the tracking database as Duplicate of INODBXXXXXX.

2.8.2.2.2.2 If not being retained as a quality assurance sample, the sample information sheet shall be marked with the original INODB number for the inmate and filed with the original form. The sample shall be destroyed.

2.8.2.3 The DNA sample(s), envelope(s) and Sample Information Sheet shall all be labeled with the corresponding INODB number.

2.8.2.4 Samples shall be stored in a secure location until analysis.

2.8.2.5 Offender samples not used for analysis shall remain in storage, indefinitely, for future reference.

2.8.3 Sample and Information Access

2.8.3.1 Per the Indiana DNA Data Base Law, access to DNA samples and DNA analysis results is limited to criminal justice agencies for law enforcement identification purposes, defense counsel for criminal defense purposes, upon authorization by a court or statute, for a population statistics database, identification research and protocol development, or quality

control purposes, but only if personal identifying information is removed. Federal, state and local law enforcement agencies may also have access through their servicing forensic DNA laboratories.

2.8.3.2 All requests for access to DNA samples or DNA analysis results except for requests from ISP Laboratories and the Indianapolis-Marion County Forensic Services Agency must be submitted in the form of a written request to the Director of the ISPL, CODIS State Administrator or CODIS Unit Supervisor. The request must identify the requesting agency, the purpose for the request and the specific data requested. Only the Laboratory Director, CODIS State Administrator or CODIS Unit Supervisor may approve such requests.

2.8.3.3 When a request for access to a DNA sample is approved, a portion of the convicted offender sample shall be released. The ISPL personnel releasing the sample shall complete an Indiana State Police Convicted Offender Sample Release Form.

2.8.3.4 When convicted offender DNA profile information is released, the ISPL personnel releasing the information shall complete a DNA Database Information Release Form.

2.8.4 Use of Contract Laboratories for analysis of Convicted Offender samples

2.8.4.1 A portion of the offender sample shall be removed from storage for analysis. The rest of the sample shall remain in storage for future reference.

2.8.4.2 Samples shall be shipped by a carrier that uses a package tracking system.

2.8.4.3 The contract laboratory shall follow sample storage procedures, analytical methods and administrative policies approved by the ISPL.

2.8.4.4 The sample analysis results shall be returned to the ISPL in an electronic format suitable for technical review.

2.8.4.5 Review of Data

2.8.4.5.1 All data from convicted offender samples received from the contract laboratory shall be reviewed by ISPL personnel that have been trained and competency tested to interpret the data.

2.8.4.5.2 This technical review shall include verification that allele designations are correct, all extraction negative, amplification

negative and amplification positive controls performed as expected and technical specifications (as agreed upon by ISPL and the contract laboratory) are met.

2.8.4.5.3 Completion of the technical review shall be documented by handwritten initials of the reviewing analyst on each page of the profile print-out and completion of an Offender Data Technical Review Worksheet.

2.8.4.5.4 When ISPL review personnel question a sample allele assignment, a decision shall be made to classify the discrepancy as a possible clerical error or a possible analytical discrepancy.

2.8.4.5.4.1 If the questioned result is clearly due to a clerical error, the allele assignment shall be changed. A notation shall be made on the paperwork received from the contract laboratory.

2.8.4.5.4.2 If the questioned result is due to a possible analytical discrepancy, the ISPL shall reanalyze the corresponding sample retained by the ISPL, or the contract laboratory shall be requested to reanalyze the sample. Whenever possible, these samples shall be reviewed with the contract laboratory during on-site visits. The ISPL shall make the final decision regarding interpretation of the sample results. When a clear interpretation is not rendered, any affected loci shall not be entered into the DNA database.

2.8.4.6 After sample analysis results are approved, the ISPL shall give the contract laboratory an e-mail or written permission to dispose of approved samples or the samples may be returned to the ISPL for storage or destruction.

2.8.4.7 Quality Assurance

2.8.4.7.1 Random Re-analysis

2.8.4.7.1.1 Duplicate quality assurance samples shall be prepared from approximately 5% of offender samples received. These samples shall be assigned a database number different from the original sample, such that the duplicate cannot be associated with the original sample.

2.8.4.7.1.2 ISPL personnel shall ensure that original and duplicate quality assurance sample results are in agreement. CODIS search software can be used to conduct or assist with this task. Discrepancies shall be resolved as appropriate.

2.8.4.7.1.3 ISPL personnel may also re-analyze selected samples.

2.8.4.7.1.4 All data received from the contract laboratory shall be reviewed and approved by ISPL personnel as described in 2.8.4.5.

2.8.4.7.2 Quality Control (QC) samples which may be an external proficiency test purchased by the ISPL shall be sent to the contract laboratory at least once per year. All DNA analysis data shall be checked for correct results and approved by ISPL personnel.

2.8.4.7.3 ISPL personnel shall perform on-site visits of the vendor laboratory at least once, prior to the initiation of analysis, to ensure the vendor laboratory is in compliance with the Quality Assurance Standards for Forensic DNA Casework Laboratories. Subsequent yearly on-site visits may be performed by another NDIS participating laboratory with appropriate review and documentation by the ISPL Technical Leader.

2.8.5 CODIS Entry

2.8.5.1 Entry of casework profiles – follow procedures in 1.8.1.

2.8.5.2 Entry of Offender profiles shall be by import of a CMF (Common Message Format) file whenever practical.

2.8.5.3 Manual entry of Offender profiles shall be verified by a second analyst. This may be done on screen and noted on the technical review worksheet.

2.8.5.4 Upload to NDIS shall be in accordance with NDIS Operational Procedures.

2.8.6 Expungement/ Administrative Deletion of Records

2.8.6.1 A DNA profile may be removed from CODIS for the following reasons:

2.8.6.1.1 A person may request expungement of their DNA profile from CODIS on the grounds that the felony conviction upon which the

authority for including the DNA profile was based has been reversed.

2.8.6.1.1.1 The person must provide a written request for expungement and a certified copy of the court order reversing the conviction.

2.8.6.1.1.2 The appropriate court jurisdiction may be contacted for verification of court action.

2.8.6.1.2 An administrative deletion shall be initiated when it has been determined a sample is no longer lawfully permitted or appropriate for retention in the system.

2.8.6.1.2.1 This type of deletion request may be initiated by ISPL personnel, DOC personnel, county collection personnel, vendor collection personnel or the convicted offender. The offender's conviction offense may be verified before removal.

2.8.6.2 Upon receipt and verification of the legitimacy of the request for removal, the CODIS State Administrator shall ensure removal of all DNA profiles, records and identifiable information in the DNA database computer pertaining to the person with regard to the dismissed or reversed conviction.

2.8.6.3 All convicted offender samples obtained from the individual that pertained to the dismissed or reversed conviction shall be destroyed.

2.8.6.4 An Indiana State Police DNA Database Removal of Convicted Offender Sample Record shall be completed.

2.8.6.5 The NDIS Expungement Procedure shall also be followed to ensure proper expungement from NDIS when applicable.

2.8.6.6 The expungement court order, sample expungement record, NDIS expungement confirmation documents, Sample Information Sheet and any other pertinent documents shall be stored in the expungement file in a secure location when not in use.

2.8.6.7 An offender requesting expungement shall be given written notification of the action taken in regard to the request.

2.8.7 CODIS Searches

2.8.7.1 Autosearches

2.8.7.1.1 On a routine basis, as determined by a CODIS Administrator or the CODIS Unit Supervisor, Autosearcher shall be initiated to search the following indexes against each other:

	Forensic	Offender	Missing Person	Relatives of Missing Person	Unidentified Human (Remains)
Forensic	X	X	X		X
Offender	X		X		X
Missing Person	X	X			X
Relatives of Missing Person					X
Unidentified Human (Remains)	X	X	X	X	X

2.8.7.1.2 Routine autosearches shall be done at Moderate Stringency with 8 loci required to report a match. If needed, the CODIS Administrator or CODIS Unit Supervisor may change the search parameters at their discretion.

2.8.7.2 Unidentified DNA profiles from unsolved cases specified on the CODIS Information Worksheet may be target searched after technical review with the approval of the CODIS State or Local Administrator or other designated personnel. A target search should only be done in the event of a rush case or other unusual circumstance and shall be documented in the case record.

2.8.7.3 The current batch target file should be downloaded from the DNA (Fax) Search Request and Batch Target Files on the CODIS website and searched against SDIS approximately quarterly.

2.8.7.4 When a DNA profile from an unsolved case is not acceptable for NDIS upload, but it may be beneficial to have it searched by other states, the DNA (Fax) Search Request and Batch Target Files procedure may be utilized.

2.8.7.5 Staff Batch Searches

2.8.7.5.1 Profiles obtained from ISPL personnel and other personnel who could potentially have contact with DNA evidence should be entered into the Staff Batch file on the SDIS CODIS Server.

2.8.7.5.2 After addition of data into CODIS, but before it is uploaded to NDIS, the Staff Batch file should be searched against all profiles present in CODIS to ensure no profile is due to a contamination event.

2.8.7.5.3 Any matches shall be immediately reported to the CODIS Administrator and/or DNA Technical Leader and the profile shall be removed from CODIS.

2.8.7.6 Familial searches (the purposeful second search of DNA profiles to identify possible genetic relatives) cannot be performed by the Indiana State Police Laboratory.

2.8.8 Hit Confirmation and Reporting

2.8.8.1 Evaluation of Matches – follow procedures in 1.8.2.1.

2.8.8.2 SDIS Offender to Case Hits

2.8.8.2.1 The CODIS Administrator or CODIS Unit Supervisor shall ensure that the proper hit confirmation steps are performed. The Hit Confirmation Checklist shall be used to document completion of the steps.

2.8.8.2.2 If the Offender has a DOC number, the ISPL may contact the Department of Correction to verify the Offender status and release date.

2.8.8.2.3 The qualifying offense of the Offender should be verified through state or county resources. If no qualifying offense can be verified, the match shall be reported under IC 10-13-6-8f.

2.8.8.2.4 The Offender profile that was entered into CODIS shall be verified by reviewing the original DNA analysis data.

2.8.8.2.5 The case profile shall be verified by reviewing the original DNA analysis data.

2.8.8.2.6 The match shall be verified by a qualified analyst by comparing the original Offender and Case profiles.

2.8.8.2.7 The remaining Offender sample shall be re-analyzed by a qualified analyst, when possible.

2.8.8.2.8 The remaining Offender sample profile shall be compared to the case profile to verify the match.

2.8.8.2.9 The thumbprint on the Offender Sample Information Sheet shall be compared to a copy of the fingerprint card obtained from Central Records to verify the identity of the Offender sample, when possible.

2.8.8.2.10 An Interstate Identification Index request shall be initiated on the Offender. This should be documented by an email placed in the hit file.

2.8.8.2.11 After verifications are complete, the match shall be confirmed by the CODIS Unit Supervisor or CODIS Administrator.

2.8.8.2.12 A LIMS report shall be generated stating the match results. No statistics shall be included in the hit report. Hit reports shall follow the CODIS Hit Report Wording in 2.11.

2.8.8.2.13 The disposition and Source ID fields, if appropriate, shall be updated.

2.8.8.2.14 If the offender is excluded as a possible contributor to the forensic sample, confirmation shall stop and the Candidate Match shall be changed to a disposition of No Match. Identifying information of the offender shall not be released.

2.8.8.2.15 Matches involving Indiana offenders with a non ISPL case shall be reported by memo, which shall be administratively reviewed for accuracy of identifying information. This review shall be documented by the reviewer's initials on the page.

2.8.8.2.16 If the offender is determined to have no convictions which qualify the individual for inclusion in CODIS, the match shall be reported as per IC 10-13-6-10. The procedure for expungement/ administrative deletion of records shall be followed to remove the individual from CODIS.

2.8.8.3 NDIS Offender to Case Hits

2.8.8.3.1 Matches involving an Indiana Offender – follow procedures in 2.8.8.2 as appropriate.

2.8.8.3.2 Matches involving an out of state Offender – follow procedures in 1.8.2.2.2.

2.8.8.4 Case to Case Hits – follow procedures in 1.8.2.3.

2.8.8.5 In instances where it is determined that the offender is not incarcerated and there are unusual public safety concerns, the investigating agency may be given verbal notification of the match with the reminder that the hit confirmation is in process. The hit report shall not be issued until the match is confirmed.

2.8.9 Systems Operations – follow procedures in 1.8.3.

2.9 Records

2.9.1 The appropriate worksheets as contained in the Biology Section Worksheet Manual shall be used to record all procedures.

2.9.2 Documents relating to hit confirmations shall be maintained in the Hit File maintained by the CODIS Unit at the Indianapolis Regional Laboratory and/or in LIMS.

2.9.3 User documentation and backup logs shall be maintained by the CODIS Administrators.

2.9.4 Sample Information Sheets, Delivery Receipts, vendor data and all other documentation relating to the collection and analysis of Offender samples shall be retained in a secure location for at least 50 years.

2.10 Interpretations of Results: All Candidate Matches shall be reviewed and evaluated by a DNA analyst currently or previously qualified in the technology being reviewed.

Matches shall be given a disposition in accordance with the disposition definitions provided in current NDIS Operational Procedures.

2.11 Report Writing

2.11.1 General rules:

2.11.1.1 Known aliases of the offender should be reported after the true name in parentheses.

2.11.1.2 One or more additional identifiers shall be included for offenders. In general, DOC# or FBI# are the most specific and beneficial, however SID#, DOB#, SSN# or any other identifiers provided may be included at the analyst's discretion.

2.11.1.3 Cases that were outsourced or analyzed in previous amplification kits shall adjust the opening paragraph accordingly.

2.11.2 Wording of CODIS Hit Results/Opinions/Interpretations: {optional}

Because the markers shall not be listed in the body of the DNA Analysis report, the following introductory statement shall precede the DNA Analysis section of a report:

In the DNA analysis detailed below, the following STR loci were analyzed by Polymerase Chain Reaction (PCR): D3S1358, TH01, D21S11, D18S51, Penta E, D5S818, D13S317, D7S820, D16S539, CSF1PO, Penta D, vWA, D8S1179, TPOX, FGA, and Amelogenin.

Note: If other markers were analyzed, they should also be listed.

Offender Hit with a Forensic Unknown (single source profile)

This report is to inform you of a potential investigative lead. {A portion of} The {partial} DNA profile obtained from the shirt (item X) was searched in the Indiana DNA Database and was found to be consistent with the convicted offender sample from John Doe (Department of Correction Inmate #123456).

If criminal prosecution is desired, please submit an evidentiary DNA standard (such as a blood standard in a purple top tube or an oral swab) from John Doe.

Offender Hit with a Forensic Mixture (or partial profile at the analyst's discretion)

This report is to inform you of a potential investigative lead. Based on a search of the Indiana DNA Database, the convicted offender sample from John Doe (Department of Correction Inmate #123456) could be a possible contributor to the DNA profile obtained from the swab (item X).

If criminal prosecution is desired, please submit an evidentiary DNA standard (such as a blood standard in a purple top tube or an oral swab) from John Doe.

Forensic Hit

This report is to inform you of a potential investigative lead. Based on a search of the Indiana DNA Database, the DNA profile obtained from the swab (item X) and the DNA profile obtained from the shirt (item X, Indiana State Police Laboratory case XX-XXXX, City Police Department case XX-XXXX) could have originated from the same individual. Please contact Detective Smith at (123) 456-7890 for more information.

Retention Notification

All sub-items created from originally submitted items will be retained by the Indiana State Police Laboratory for the possibility of future analysis.

If No or Insufficient Print

Please note that fingerprint confirmation of the offender's identity was not possible in this case.

2.12 References: None

3 ISPL Analysis of Convicted Offender Samples

3.1 Scope:

This test method is designed for the guidance of Laboratory personnel who develop profiles from samples of convicted offenders collected for inclusion in the CODIS database. Samples may include blood stain cards or buccal/oral swabs. This manual may be expanded or altered as new techniques and/or genetic systems are available with the approval of the Division Commander.

3.1.1 The DNA is incubated, directly amplified to produce many fluorescently tagged copies of specific regions of the DNA and processed to separate and detect a DNA profile. These profiles are then entered into CODIS.

3.2 Precautions/Limitations:

3.2.1 Convicted Offender samples collected for Combined DNA Index System (CODIS) shall be treated as reference materials and not considered evidence.

3.3 Related Information:

3.3.1 Worksheet Manual

3.4 Instruments:

3.4.1 Applied Biosystems™ 3500xl Genetic Analyzer Capillary Electrophoresis Instrument – Simultaneously separates and detects multiple amplified DNA samples by size by capillary electrophoresis using fluorescent tagged primers.

3.4.2 Laminar Flow Hood – An air purifying biohazard cabinet that maintains a nominal inflow velocity of 80 fpm which prevents contaminants from entering or escaping the work area.

3.4.3 Miscellaneous Laboratory Equipment - Supportive laboratory equipment consisting of ovens, incubators, minifuges, pipettes, water baths, stirring/heating plates, vortex mixers, thermometers, temperature verification system, vacuum pump, refrigerators/freezers for storing of reagents, buffers and work product.

3.4.4 Thermal cycler - An instrument that can be programmed to rapidly cycle between high and low temperatures. This process is used to make many fluorescently tagged copies of specific regions on a DNA strand(s).

3.4.5 Water Purification System - An apparatus that routes water through a series of filtering devices to produce high quality, uncontaminated water used in buffer preparation and DNA typing methods.

3.5 Reagents/Materials: Reagents critical to the DNA analysis process are listed in the Critical Reagent Manual.

3.5.1 Proteinase K (Pro K) 18mg/ml
3.5.2 Stain Extraction Buffer for Automation
3.5.3 Nuclease Free Water (NFH2O)

3.6 Hazards/Safety:

3.6.1 All chemicals shall be handled in a safe method as referenced in the specific Material Safety Data Sheets (MSDS).

3.6.2 The manual preparation of samples for electrophoresis by the addition of Hi-Di™ formamide shall be confined to a chemical fume hood. **Caution**: Formamide is an irritant and teratogen; therefore universal precautions and a fume hood shall be utilized when manually working with formamide to avoid inhalation and contact with the skin.

3.6.3 Universal Precautions shall be in use whenever biological materials are being handled.

3.6.4 Biological waste shall be disposed of in the appropriate waste receptacle.

3.7 Reference Materials/Controls/Calibration Checks:

3.7.1 DNA profiles shall be developed in compliance with the DNA Identification Act of 1994, the FBI Approved Quality Assurance Standards for Forensic DNA Testing Laboratories and the FBI Approved Quality Assurance Standards for DNA Databasing Laboratories.

3.7.2 The accuracy and specificity of test results are ensured by running known DNA controls and reagent controls at the same time as samples. See the specific test for the appropriate controls to be run and the interpretation of the results.

3.8 Procedures/Instructions:

3.8.1 PowerPlex® 18D Introduction (Applied Biosystems® 3500xl Genetic Analyzer)

3.8.1.1 The Promega PowerPlex® 18D System allows for the amplification of seventeen short tandem repeat (STR) loci and the Amelogenin locus found on the X and Y chromosomes (see chart on next page). The amplification occurs in a single reaction tube and detection occurs by a single capillary electrophoresis injection. The overlapping loci can be visualized simultaneously by using PCR primers labeled with four different fluorescent tags (see chart on next page).

3.8.1.2 The Applied Biosystems® 3500xl Genetic Analyzer utilizes electrokinetic injection of DNA molecules into polymer-filled capillaries which separates the DNA fragments by size. The fluorescent tag labeled primers incorporated into the PowerPlex® 18D amplification products are responsive to the frequency of the 505nm solid state laser. Upon excitation, the fluorophores are raised to a higher energy level. When the fluorophores return to their normal energy level, a fluorescent signal is emitted. This signal is then detected by a camera within the 3500xl capillary electrophoresis instrument which converts the signal to a computer image where it is visualized in an electropherogram as a peak.

3.8.1.3 The data produced by the 3500xl Genetic Analyzer is analyzed with GeneMapper® *ID*-X Software which results in peaks labeled with their allele designation. The allele designation for each sample is accomplished through the use of an internal lane standard (ILS). The ILS is injected with each sample and it contains 21 fragments of known length. The ILS determines the base pair size of the fragments in the sample and the software compares the sizes to an allelic ladder to determine the allele designation.

INDIANA STATE POLICE
BIOLOGY SECTION
DATABASING TEST METHODS

The PowerPlex® 18D System Locus-Specific and Allelic Ladder Information

Locus	Chromosomal Location	Repeat Sequence[1] 5'-->3'	Allelic Ladder Size Ranges [3,4] (bases)	STR Ladder Alleles[5] (# of repeats)	Fluorophore
Penta E	15q	AAAGA*	379-474	5-24	FL
D18S51	18q21.3	AGAA*	286-366	7-10, 10.2, 11-13, 13.2, 14-27	FL
D21S11	21q11-21q21	TCTA*	203-259	24, 24.2, 25, 25.2, 26-28, 28.2, 29, 29.2, 30, 30.2, 31, 31.2, 32, 32.2, 33, 33.2, 34, 34.2, 35, 35.2, 36-38	FL
TH01	11p15.5	AATG*	152-195	3-9, 9.3, 10-11, 13.3	FL
D3S1358	3p	TCTA*	103-147	9-20	FL
FGA	4q28	TTTC*	314-460	14-18, 18.2, 19, 19.2, 20, 20.2, 21, 21.2, 22, 22.2, 23, 23.2, 24, 24.2, 25, 25.2, 26-30, 31.2, 32.2, 33.2, 42.2, 43.2, 44.2, 45.2, 46.2, 48.2, 50.2	TMR-ET
TPOX	2p24-2pter	AATG*	265-293	6-13	TMR-ET
D8S1179	8q	TCTA*	203-251	7-19	TMR-ET
vWA	12p12-pter	TCTA*	127-183	10-24	TMR-ET
Amelogenin[2]	Xp22.1-22.3 and Y	N/A	109(X)/115(Y)	X,Y	TMR-ET
Penta D	21q	AAAGA*	376-449	2.2, 3.2, 5-17	JOE
CSF1PO	5q33.3-34	AGAT*	321-357	6-15	JOE
D16S539	16q24-qter	GATA*	264-304	5,8-15	JOE
D7S820	7q11.21-22	GATA*	218-250	6-14	JOE
D13S317	13q22-q31	TATC*	176-208	7-15	JOE
D5S818	5q23.3-32	AGAT*	122-158	7-16	JOE
D2S1338	2q35	TGCC/TTCC	223-295	10, 12, 14-28	CXR-ET
D19S433	19q12	AAGG	163-215	5.2, 6.2, 8-12, 12.2, 13, 13.2, 14, 14.2, 15, 15.2, 16, 16.2, 17, 17.2, 18, 18.2	CXR-ET

[1] The August 1997 report (25,26) of the DNA Commission of the International Society for Forensic Haemogenetics (ISFH) states, "1) for STR loci within coding genes, the coding strand shall be used and the repeat sequence motif defined using the first possible 5´ nucleotide of a repeat motif; and 2) for STR loci not associated with a coding gene, the first database entry or original literature description shall be used.".

[2] Amelogenin is not an STR but displays a 109-base, X-specific band and a 115-base, Y-specific band. 9947A DNA (female) displays only the 109-base, X-specific band.

[3] The length of each allele in the allelic ladder has been confirmed by sequence analyses.

[4] When using an internal lane standard, such as the CC5 Internal Lane Standard 500, the calculated sizes of allelic ladder components may differ from those listed. This occurs because different sequences in allelic ladder and ILS components may cause differences in migration. The dye label also affects migration of alleles.

[5] For a current list of microvariants, see the Variant Allele Report published at the U.S. National Institute of Standards and Technology (NIST) web site at: **www.cstl.nist.gov/div831/strbase/**

NA = not applicable

3.8.2 PowerPlex® 18D Amplification

3.8.2.1 When programming the GeneAmp® PCR System 9700 thermal cycler, use the ramping mode for the GeneAmp® PCR System 9600 thermal cycler.

3.8.2.2 Select 25µl for the volume.

3.8.2.3 Applied Biosystems GeneAmp® 9700 Thermal Cycler Program

3.8.2.3.1 96°C for 2 minutes, then:

3.8.2.3.2 94°C for 10 seconds

3.8.2.3.3 60°C for 60 seconds

3.8.2.3.4 for 27 cycles, then:

3.8.2.3.5 60°C for 20 minutes, then:

3.8.2.3.6 Soak at 4°C until the plate is removed.

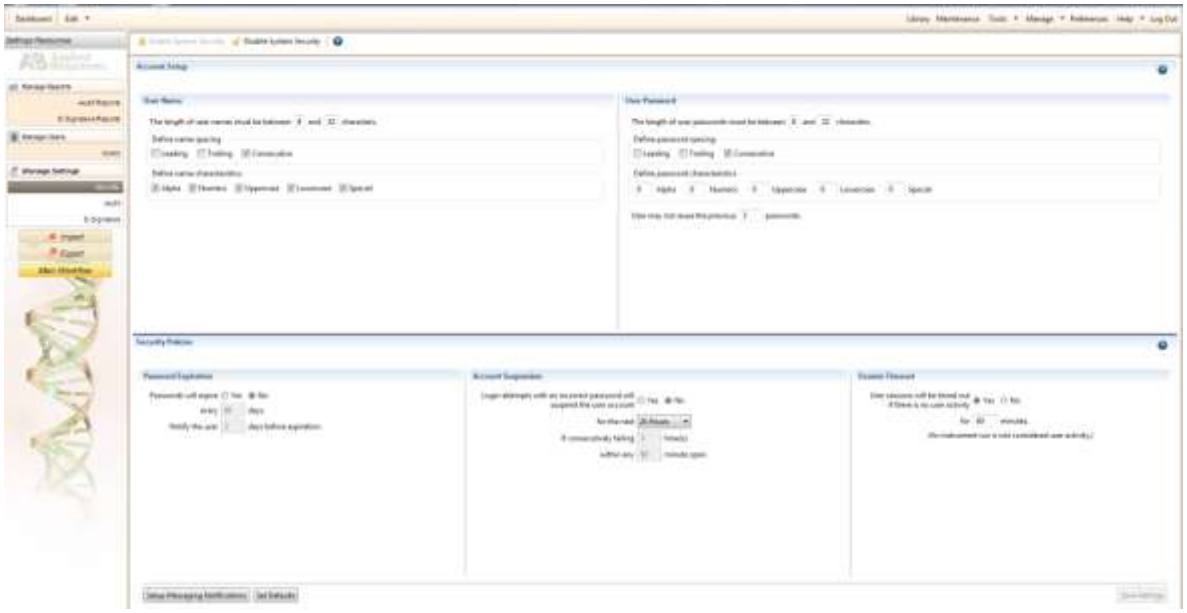
3.8.3 Applied Biosystems 3500xl Genetic Analyzer - Data Collection Software version 2.0 - Instrument Set-up

3.8.3.1 Configure the Security Settings

3.8.3.1.1 Navigate to Tools and select “Security”.

3.8.3.1.2 Change Screen settings to those depicted below.

Security Screen



3.8.3.1.3 Click “Save Settings” button.

3.8.3.2 Edit User Roles

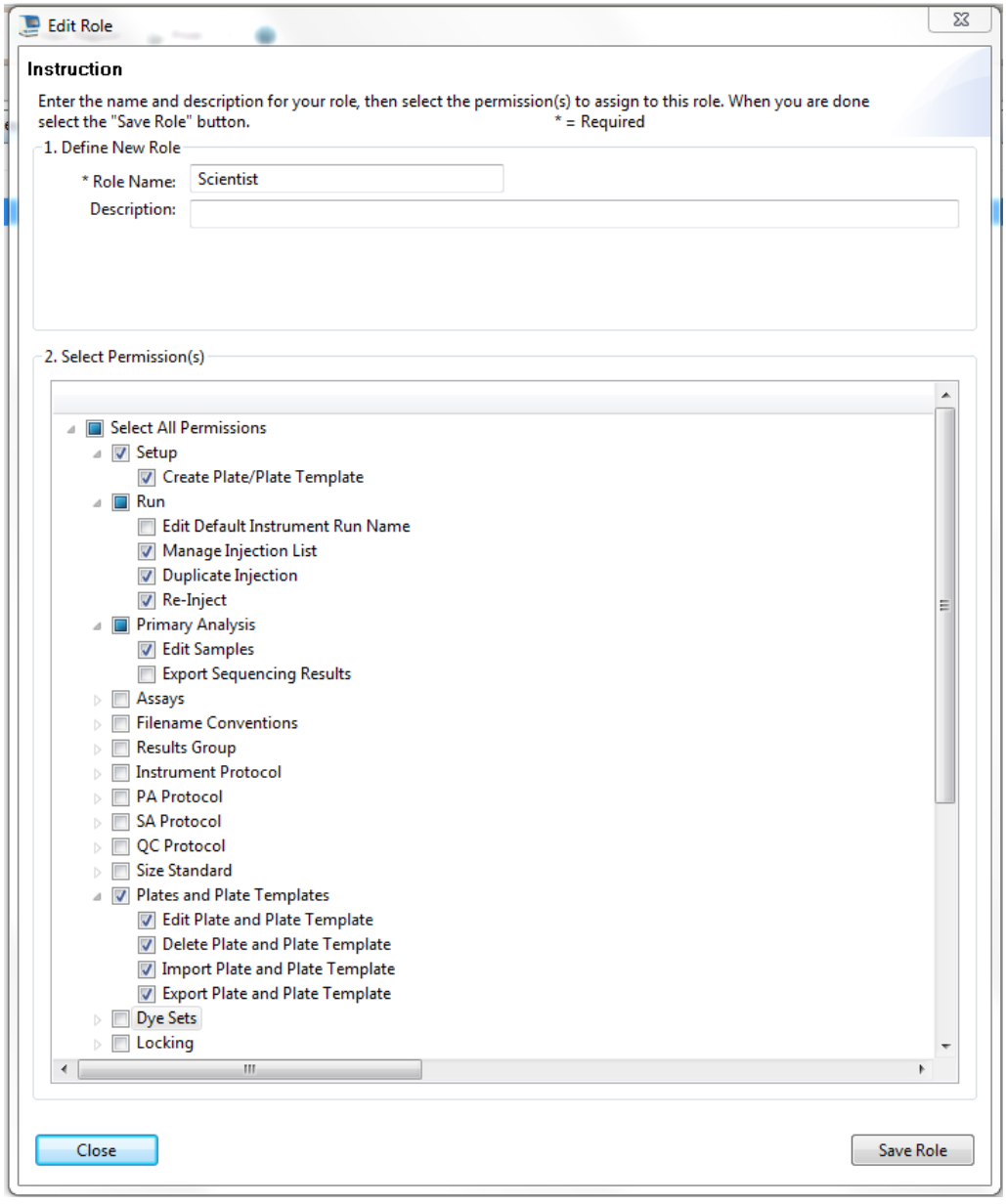
3.8.3.2.1 Click the “Users” button in the left navigation pane.

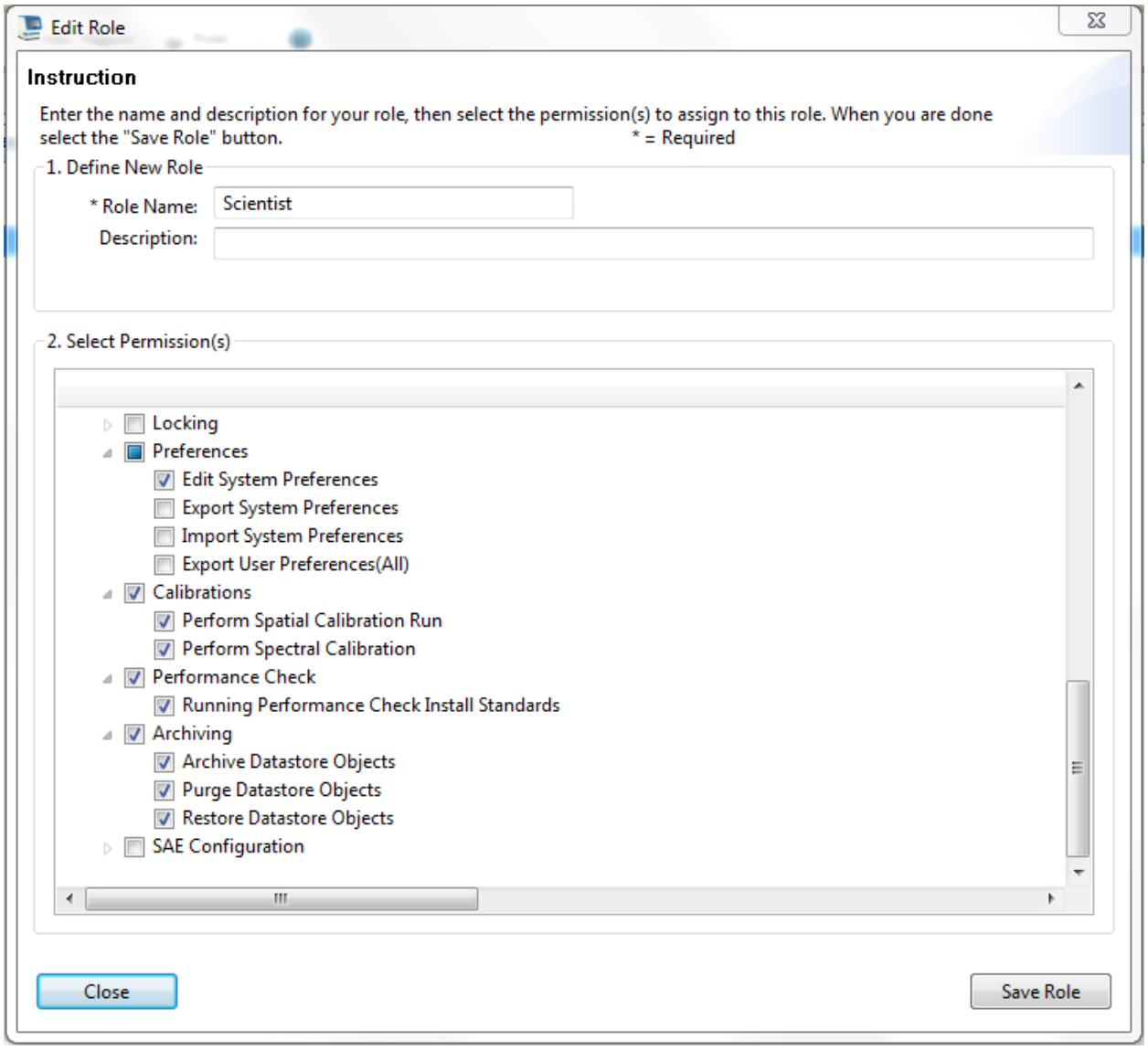
3.8.3.2.2 Click the “Roles” tab.

3.8.3.2.3 Set preferences to those depicted below.

3.8.3.2.4 Select “Scientist” and click “Edit”.

Scientist User Role





3.8.3.2.5 Click “Save Role”.

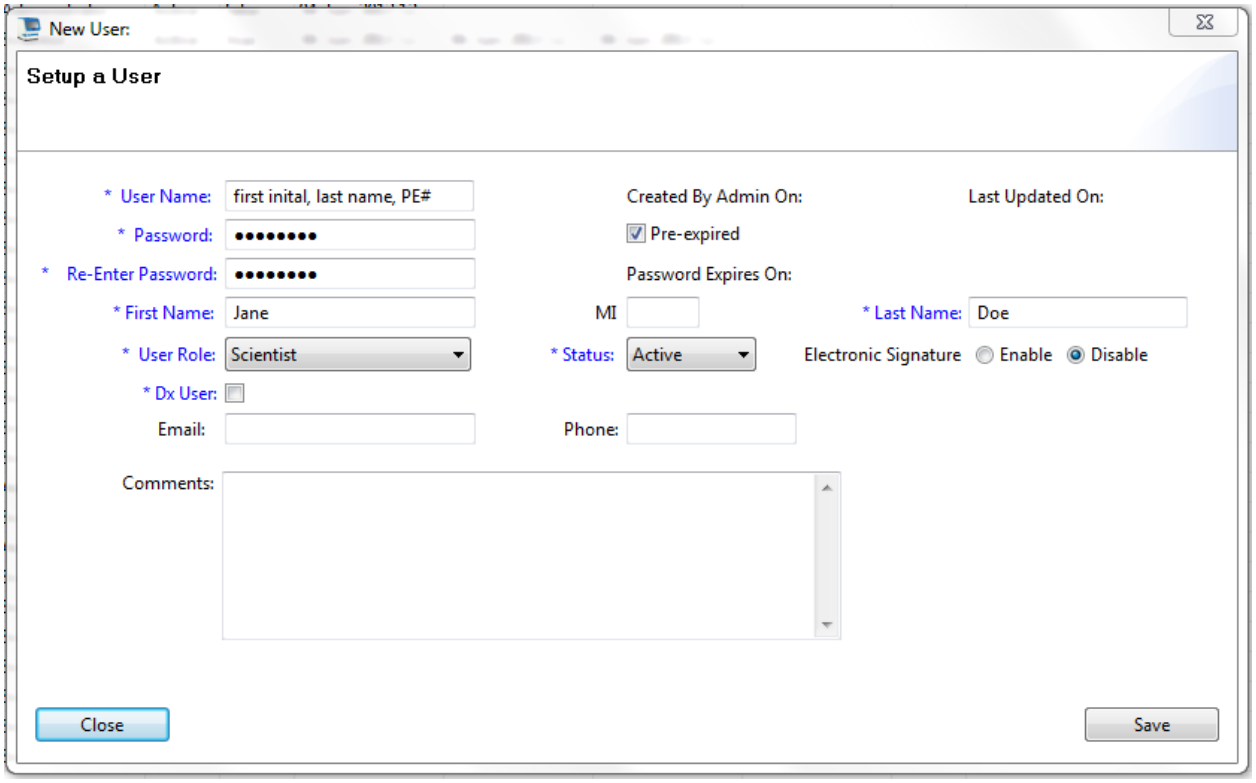
3.8.3.3 Create User Accounts

3.8.3.3.1 Click on the “Users” tab.

3.8.3.3.2 Click “Create” to access a New User window.

3.8.3.3.3 Enter a unique User Name (ex. jdoe8251), set “Password” to lowercase “password” and re-enter. Enter user’s first and last name and change settings to those depicted below. Ensure the “User Role” is either set as an “Administrator” or “Scientist”.

New User Window



3.8.3.3.4 Click the “Save” button.

3.8.3.3.5 Repeat the above steps to create a User Account for each analyst.

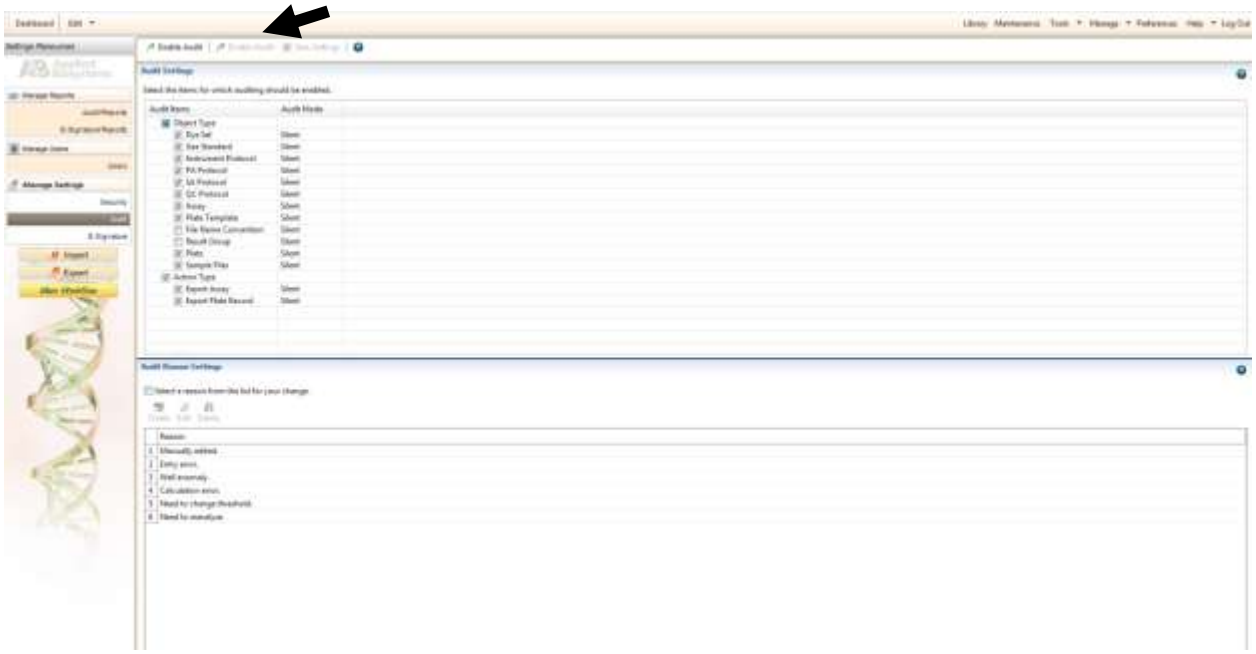
3.8.3.4 Manage Audit Settings

3.8.3.4.1 Navigate to Tools and Select “Audit”.

3.8.3.4.2 Turn Auditing off by ensuring the “Disable Audit” is grayed out and the “Enable Audit” appears black as depicted below.

3.8.3.4.3 Audit settings may be altered depending on current need.

Disable Audit Settings



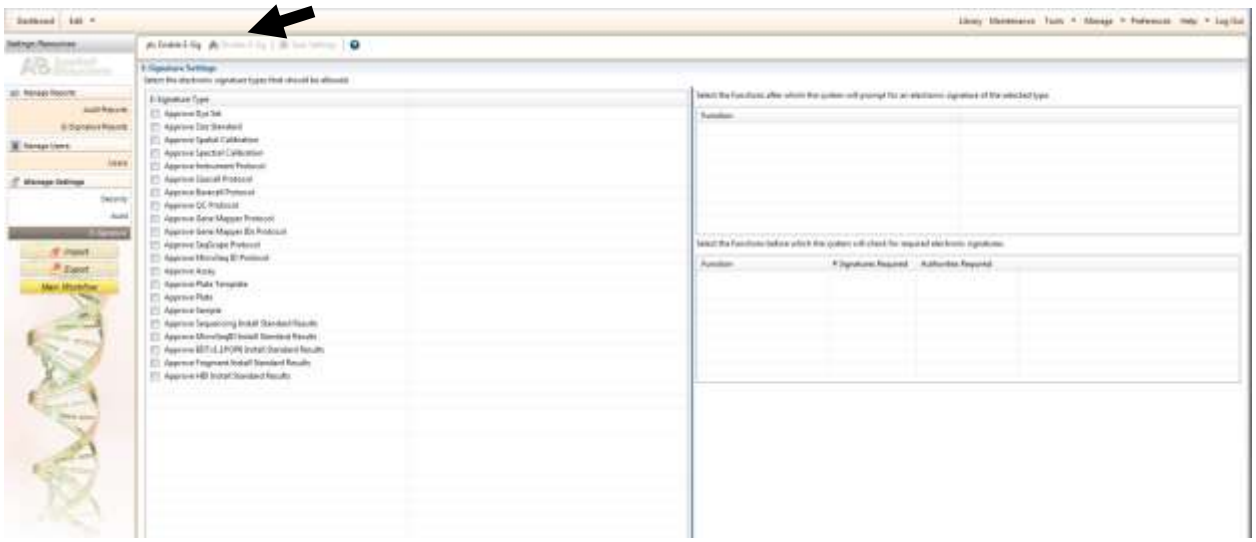
3.8.3.5 Manage Electronic Signature Settings

3.8.3.5.1 Navigate to Tools and Select “E-Signature”.

3.8.3.5.2 Turn E-Signature off by ensuring that the “Disable E-Sig” is grayed out and that the “Enable E-sig” is black as depicted below.

3.8.3.5.3 E-Signature settings may be altered depending on need.

Disable E-Signature Settings



3.8.3.6 Create an Instrument Protocol

3.8.3.6.1 Navigate to the Library and Select “Instrument Protocols”.

3.8.3.6.2 Select “Create”.

3.8.3.6.3 Select “HID” for the Application Type.

3.8.3.6.4 Select “36” for the Capillary Length.

3.8.3.6.5 Select “POP4” for the Polymer.

3.8.3.6.6 Select “PromegaG5” for the Dye Set.

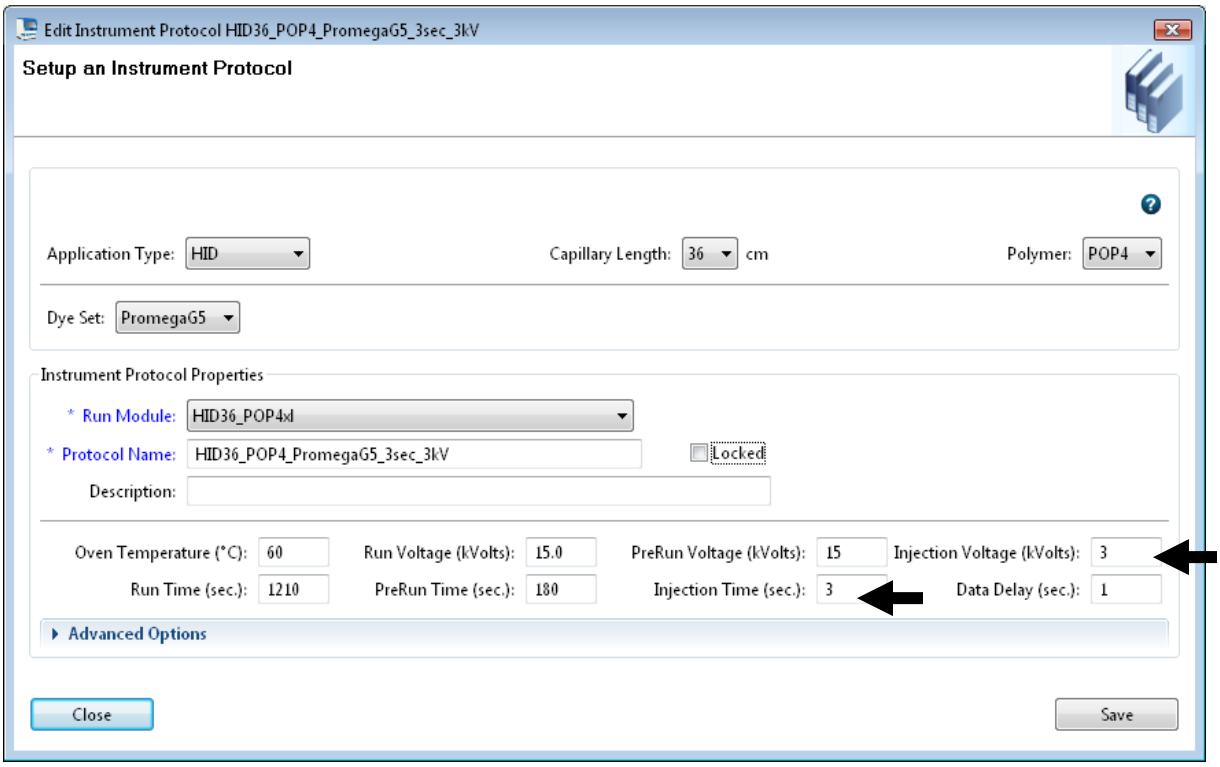
3.8.3.6.7 Select “HID36_POP4xl” for the Run Module.

3.8.3.6.8 Select the desired injection time and injection voltage.

3.8.3.6.9 Leave all other settings as default.

3.8.3.6.10 Name the protocol with a descriptive name (ex. HID36_POP4_Promega G5_3sec_3kV) and select “Save”.

Instrument Protocol



3.8.3.6.11 Instrument protocols shall be created for 1kV/5sec, 3kV/3sec, 3kV/5sec and 3kV/8sec.

3.8.3.7 Create a size standard

3.8.3.7.1 Navigate to the Library and Select “Size Standard”.

3.8.3.7.2 Select “Create”.

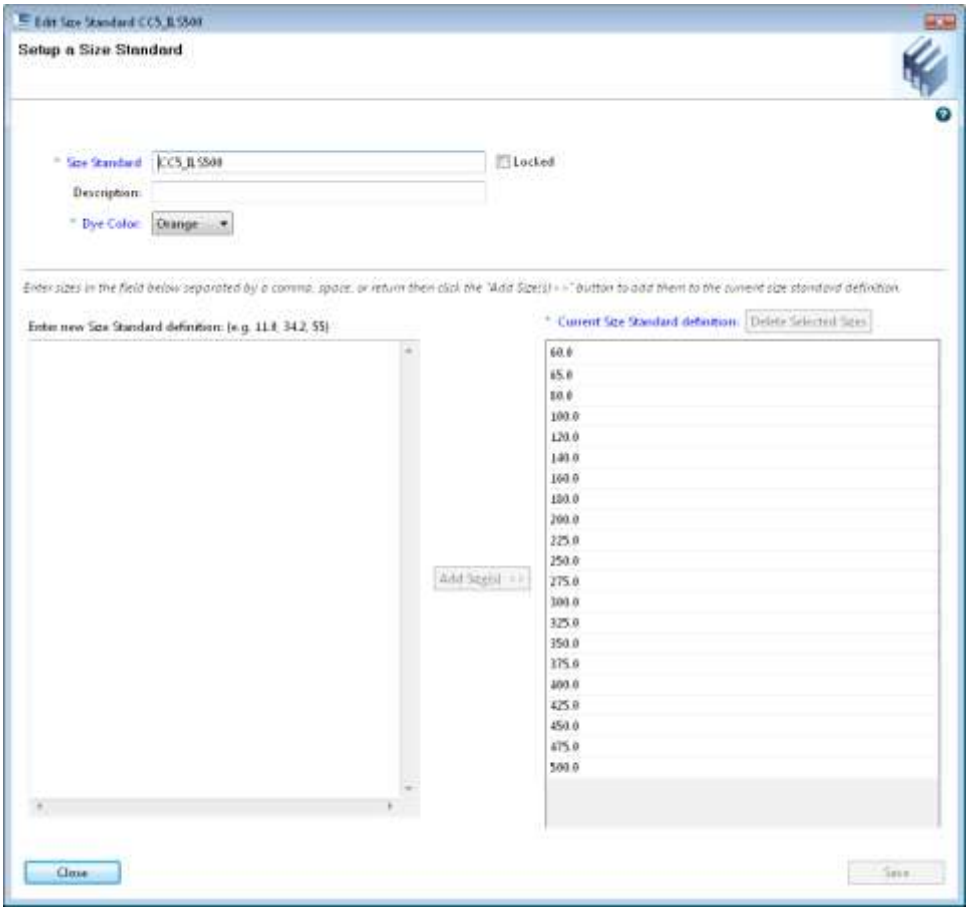
3.8.3.7.3 Assign the name “CC5_ILS500”.

3.8.3.7.4 Select “Orange” for the Dye Color.

3.8.3.7.5 Type in the following values: 60, 65, 80, 100, 120, 140, 160, 180, 200, 225, 250, 275, 300, 325, 350, 375, 400, 425, 450, 475 and 500 and click “Add Size(s)”.

3.8.3.7.6 Click “Save”.

Size Standard



3.8.3.8 Create a QC Protocol

3.8.3.8.1 Navigate to the Library and Select “QC Protocols”.

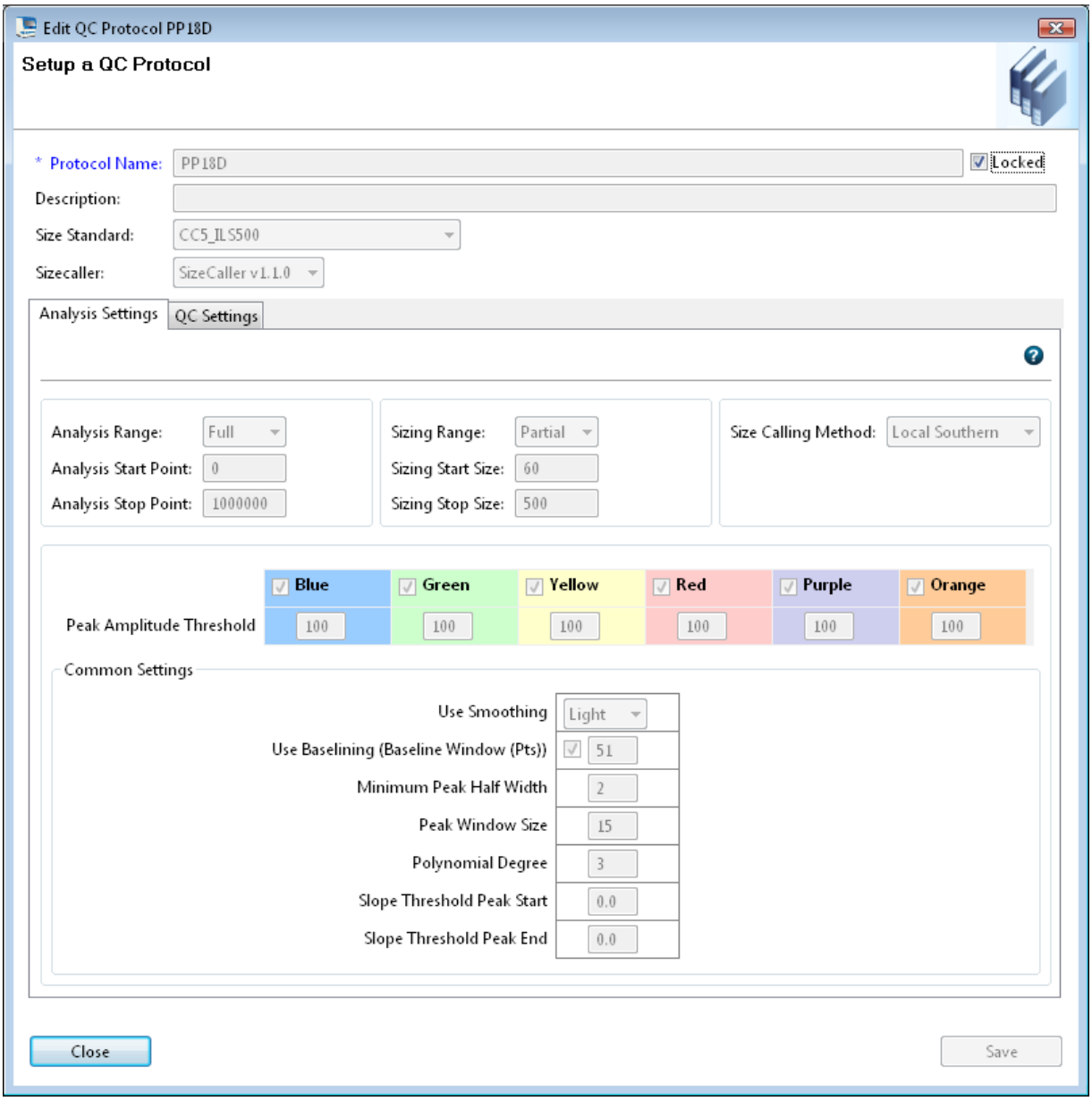
3.8.3.8.2 Select “Create”.

3.8.3.8.3 Assign the name “PP18D”.

3.8.3.8.4 Change settings to those shown below.

3.8.3.8.5 Click “Save”.

QC Protocol



3.8.3.9 Create an Assay

3.8.3.9.1 Navigate to the Library and Select “Assays”.

3.8.3.9.2 Select “Create”.

3.8.3.9.3 Assign a descriptive assay name (ex. PP18D_3kV3sec).

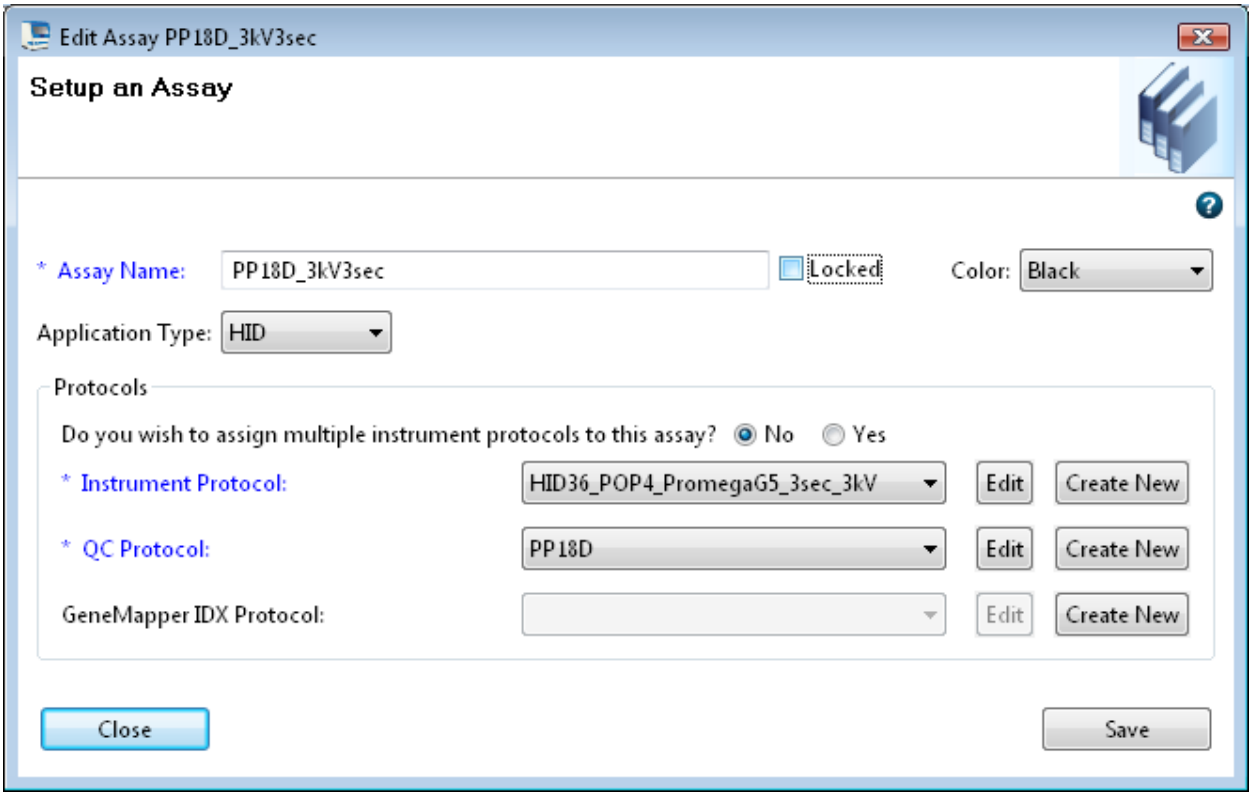
3.8.3.9.4 Select “HID” for the Application Type.

3.8.3.9.5 Select “PP18D” as the QC Protocol.

3.8.3.9.6 Select the appropriate instrument protocol created in 3.8.3.1.

3.8.3.9.7 Click “Save”.

Assay



3.8.3.9.8 An assay shall be created for each instrument protocol.

3.8.3.9.9 If multiple instrument protocols need to be run on the same samples, an assay shall be created for that combination of protocols.

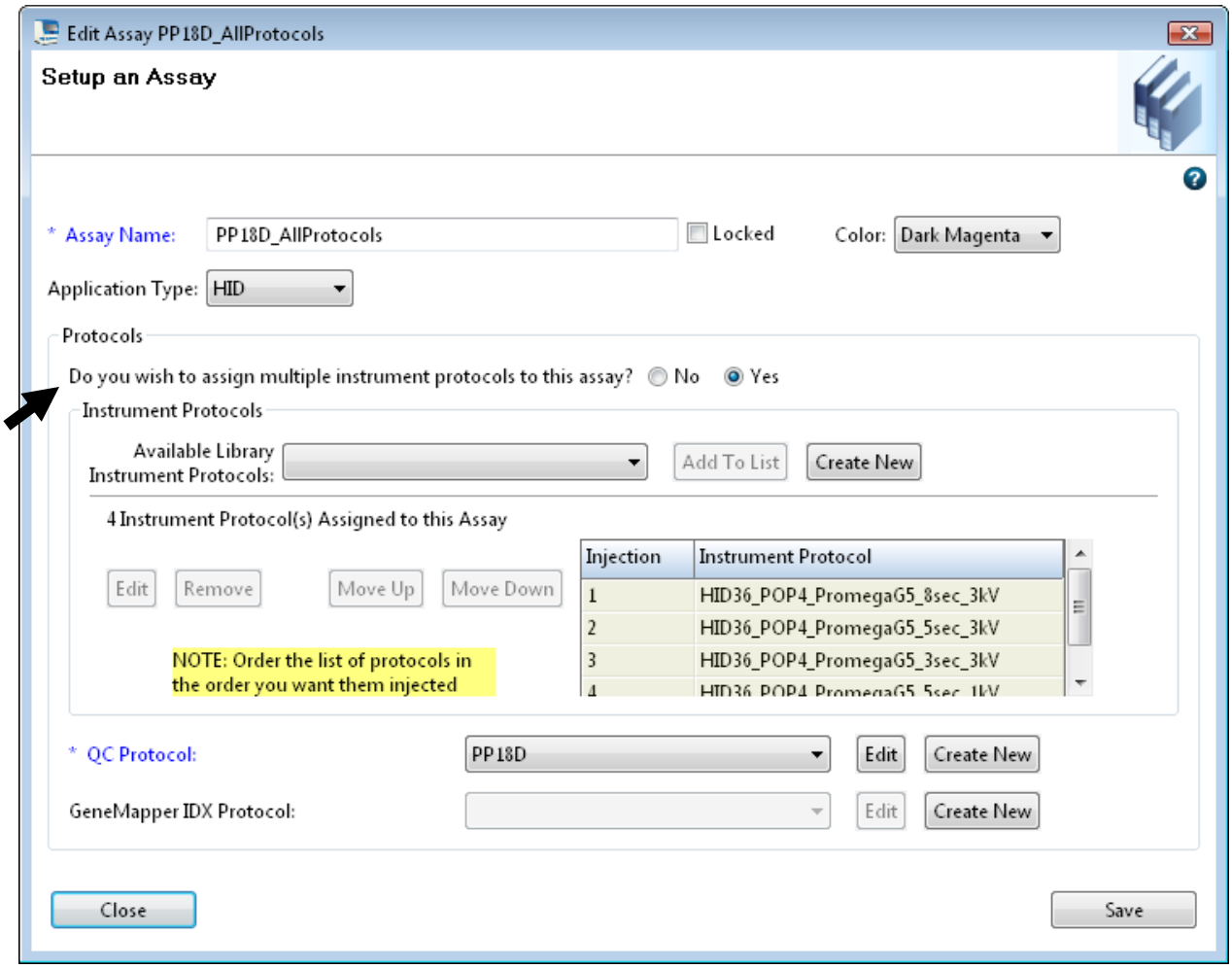
3.8.3.9.9.1 Follow steps 3.8.3.4.1 through 3.8.3.4.6.

3.8.3.9.9.2 Select “Yes” to assign multiple instrument protocols to this assay.

3.8.3.9.9.3 Select each desired instrument protocol and click Add to List.

3.8.3.9.9.4 Click “Save”.

Assay: Multiple Injections



3.8.3.10 Create a Naming Convention

3.8.3.10.1 Navigate to the Library and Select “File Name Convention”.

3.8.3.10.2 Select “Create”.

3.8.3.10.3 Assign a descriptive name (ex. CODIS).

3.8.3.10.4 Select desired attributes and click “Add”.

3.8.3.10.5 Select desired delimiters and click “Add”.

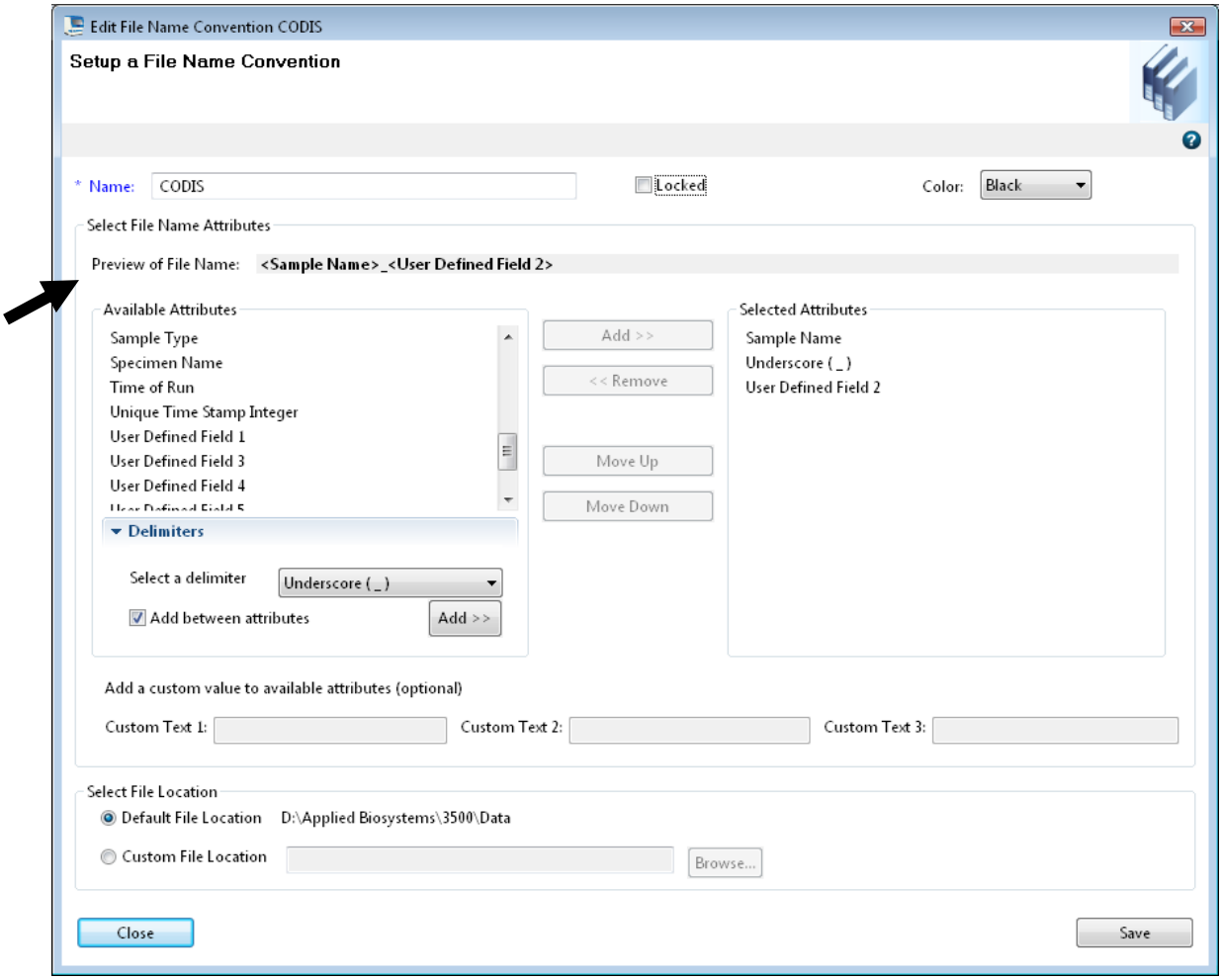
3.8.3.10.6 Click so the “Add between attributes” box is checked.

3.8.3.10.7 The order of the attributes and delimiters can be changed using the Move Up and Move Down buttons.

3.8.3.10.8 Leave file location as the default.

3.8.3.10.9 Look at the Preview of File Name field for accuracy and Click “Save”.

File Name Convention



3.8.3.10.10 For offender analysis, the File Name Convention shall be Sample Name _User Defined Field 2.

3.8.3.11 Create a Results Group

3.8.3.11.1 Navigate to the Library and Select “Results Group”.

3.8.3.11.2 Select “Create”.

3.8.3.11.3 Assign a descriptive assay name (ex. PP18D).

3.8.3.11.4 Select desired attributes and click “Add”.

3.8.3.11.5 Select desired delimiters and click “Add”.

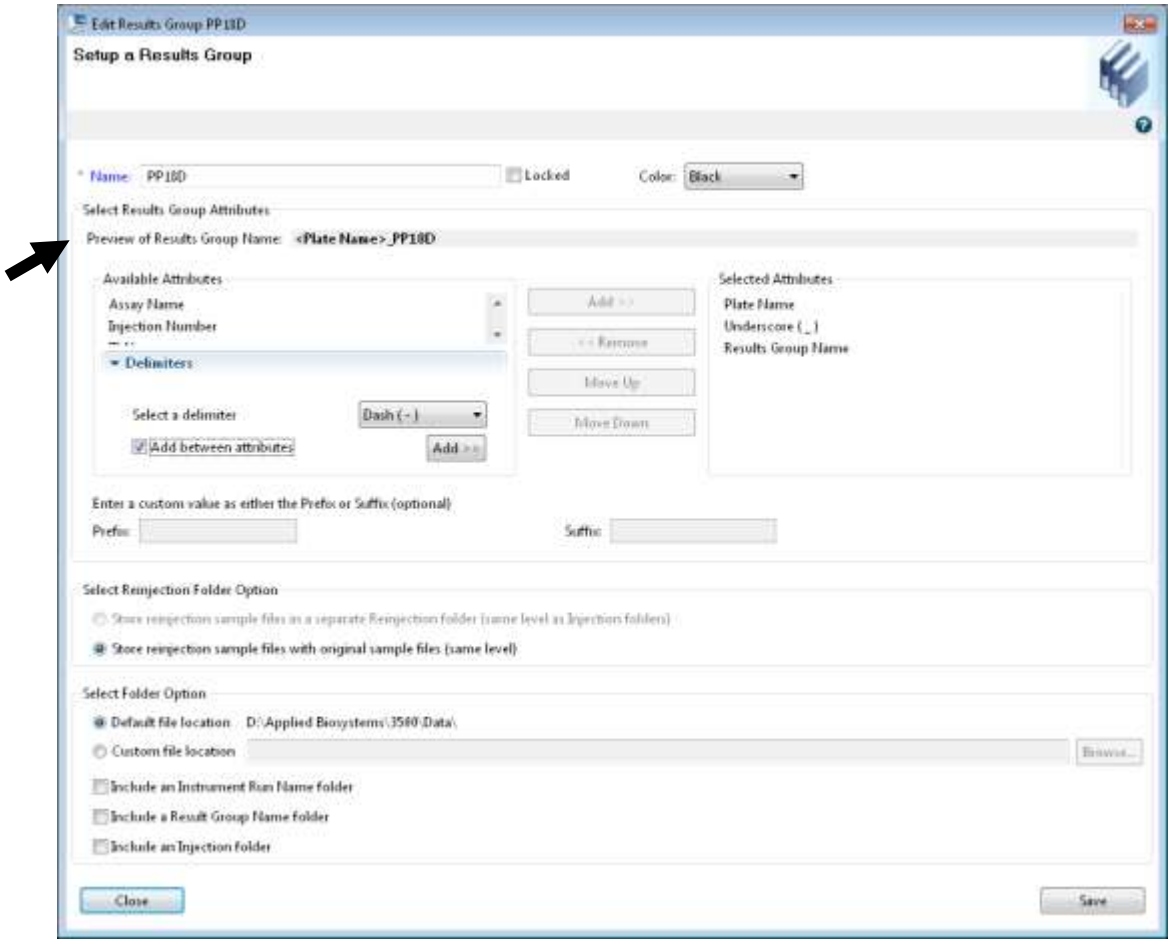
3.8.3.11.6 Click so the “Add between attributes” box is checked.

3.8.3.11.7 The order of the attributes and delimiters can be changed using the Move Up and Move Down buttons.

3.8.3.11.8 Leave file location as the default.

3.8.3.11.9 Look at the Preview of Results Group Name field for accuracy and Click "Save".

Results Group



3.8.3.11.10 For offender analysis, the Results Group Name shall be Plate Name_Results Group Name.

3.8.4 GeneMapper® *ID*-X Software version 1.4 - PowerPlex® 18D Software Settings

3.8.4.1 Import Panel and Bin Files

3.8.4.1.1 Select “Tools”, then “Panel Manager”.

3.8.4.1.2 Highlight the Panel Manager icon in the upper left navigation pane.

3.8.4.1.3 Select “File”, then “Import Panels”.

3.8.4.1.4 Navigate to the saved panels and bins file. Select “PowerPlex_18D_Panels_IDX_v1.2.txt”, then “Import”.

3.8.4.1.5 In the navigation pane, highlight “PowerPlex_18D_Panels_IDX_v1.2.txt”.

3.8.4.1.6 Select “File”, then “Import Bin Set”.

3.8.4.1.7 Navigate to the bins file. Select “PowerPlex_18D_Bins_IDX_v1.2.txt”, then “Import”.

3.8.4.1.8 In the navigation pane, highlight “PowerPlex_18D_Bins_IDX_v1.2.txt”.

3.8.4.1.9 Select “File”, then “Import Marker Stutter”. A warning box will appear asking if you want to overwrite current values. Select “Yes”.

3.8.4.1.10 Navigate to the stutter file. Select “PowerPlex_18D_Stutter_IDX_v1.2.txt”, then “Import”.

3.8.4.1.11 At the bottom of the Panel Manager window, select apply and then “OK”.

3.8.4.2 Create an Analysis Method

3.8.4.2.1 Select “Tools”, then “GeneMapper ID-X Manager”.

3.8.4.2.2 Select the “Analysis Methods” tab.

3.8.4.2.3 Select “New” and a new analysis method dialog box will open.

3.8.4.2.4 Enter the name “PP18D CODIS” for the analysis method.

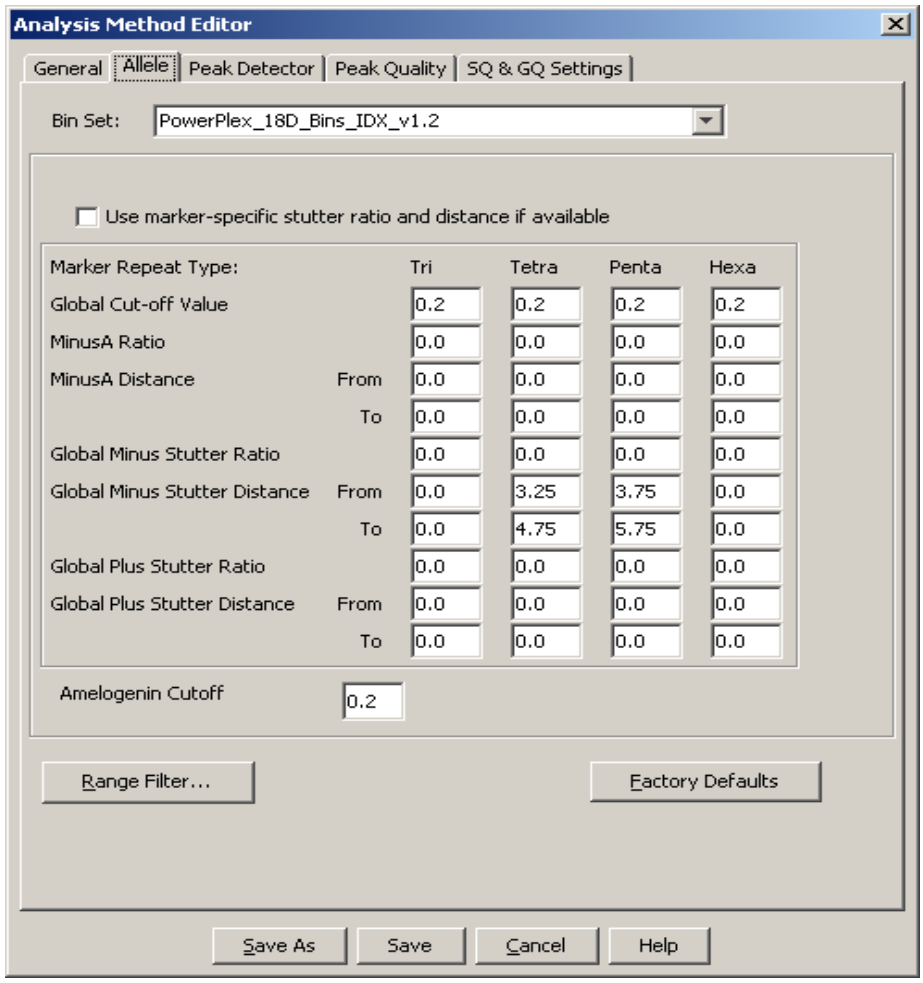
3.8.4.2.5 In the Analysis Method Editor window, select "ISP Databasing Security Group" as the Security Group.

3.8.4.2.6 Select the "Allele" tab.

3.8.4.2.7 Select "PowerPlex_18D_Bins_IDX_v1.2" as the Bin Set.

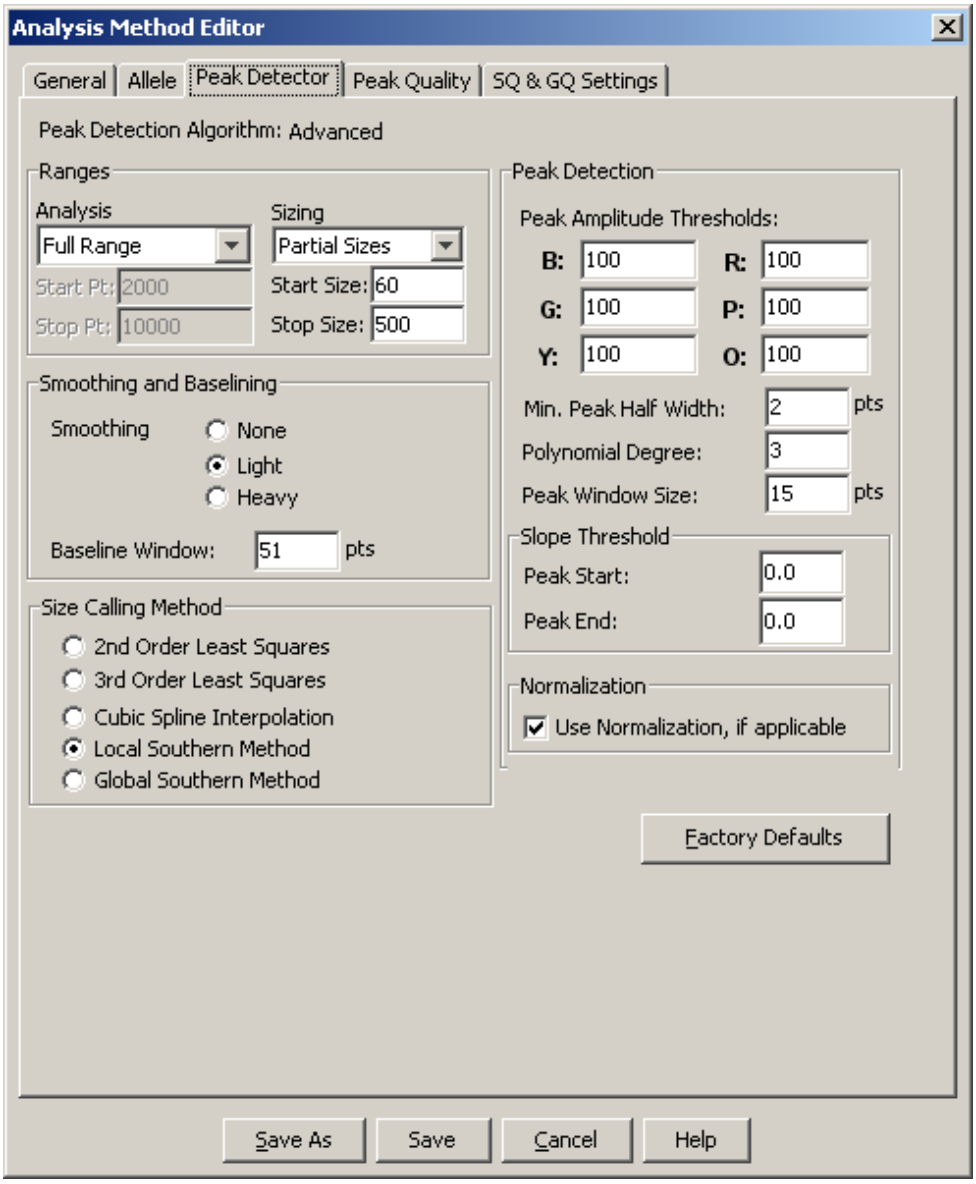
3.8.4.2.8 Ensure that the global cut-off value is set at 0.2 for all.

Allele Tab



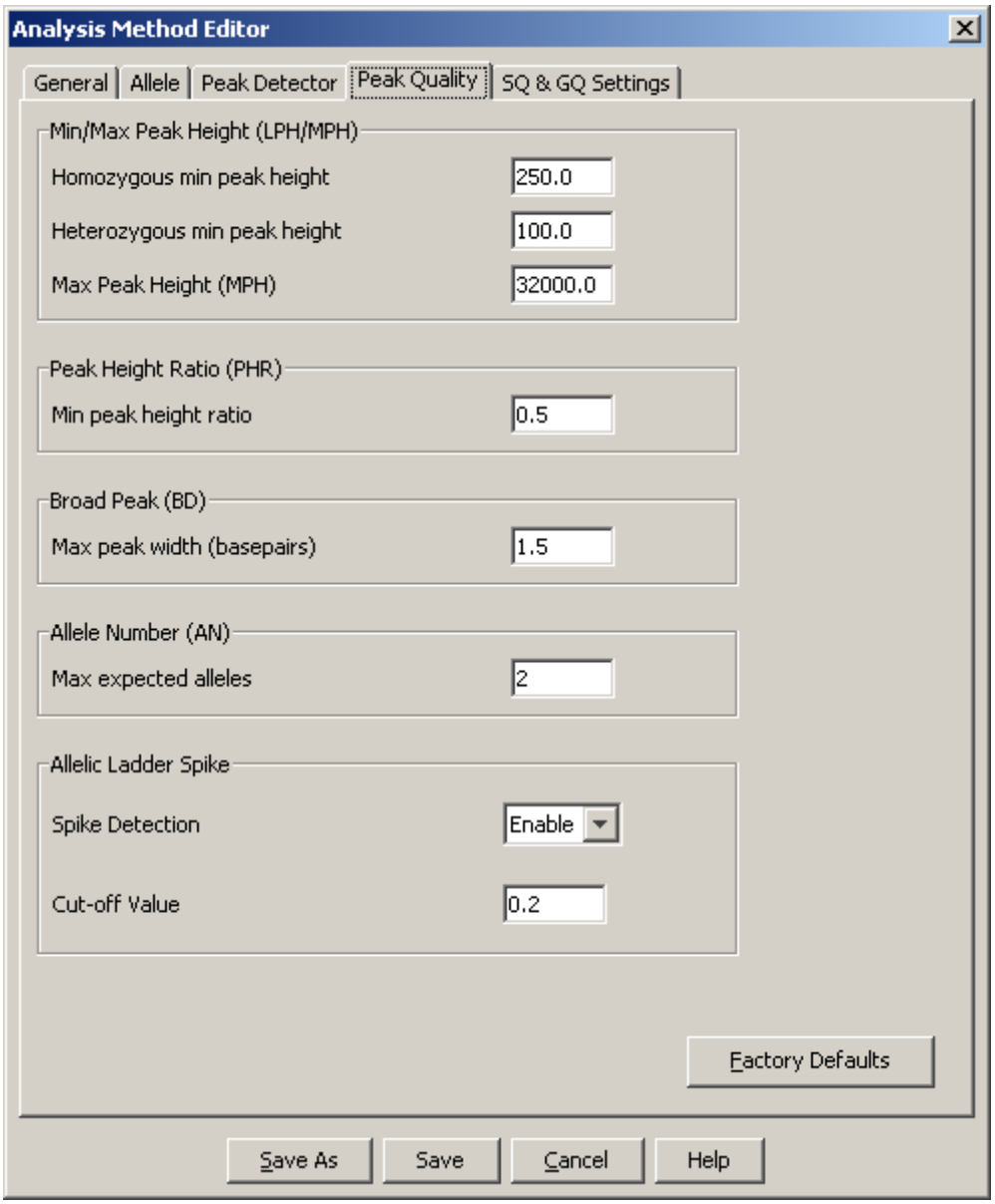
3.8.4.2.9 Select "Peak Detector" tab. Change the settings to those shown below. Alternatively, the Analysis Range may be set to "Partial Range" and the start point adjusted if the Instrument migration warrants it (to eliminate the primer peak).

Peak Detector Tab



3.8.4.2.10 Select the "Peak Quality" tab. Change settings to those shown below.

Peak Quality Tab



3.8.4.2.11 Leave all settings in the "SQ & GQ Settings" tab at factory defaults.

3.8.4.2.12 Click "Save".

3.8.4.3 Create a Size Standard

3.8.4.3.1 Select "Tools", then "GeneMapper ID-X Manager".

3.8.4.3.2 Select the "Size Standard" tab.

3.8.4.3.3 Select “New”.

3.8.4.3.4 In the Size Standard Editor window, select “ISP Databasing Security Group” as the Security Group.

3.8.4.3.5 Enter the name as “CC5_ILS500”.

3.8.4.3.6 Choose “Orange” for the Size Standard Dye.

3.8.4.3.7 Enter the sizes of the internal lane standard fragments (60, 65, 80, 100, 120,140, 160, 180, 200, 225, 250, 275, 300, 325, 350, 375, 400, 425, 450, 475 and 500 bases).

3.8.4.3.8 Click “OK”.

3.8.4.4 Create a Table Setting

3.8.4.4.1 Select “Tools”, then “GeneMapper Manager”.

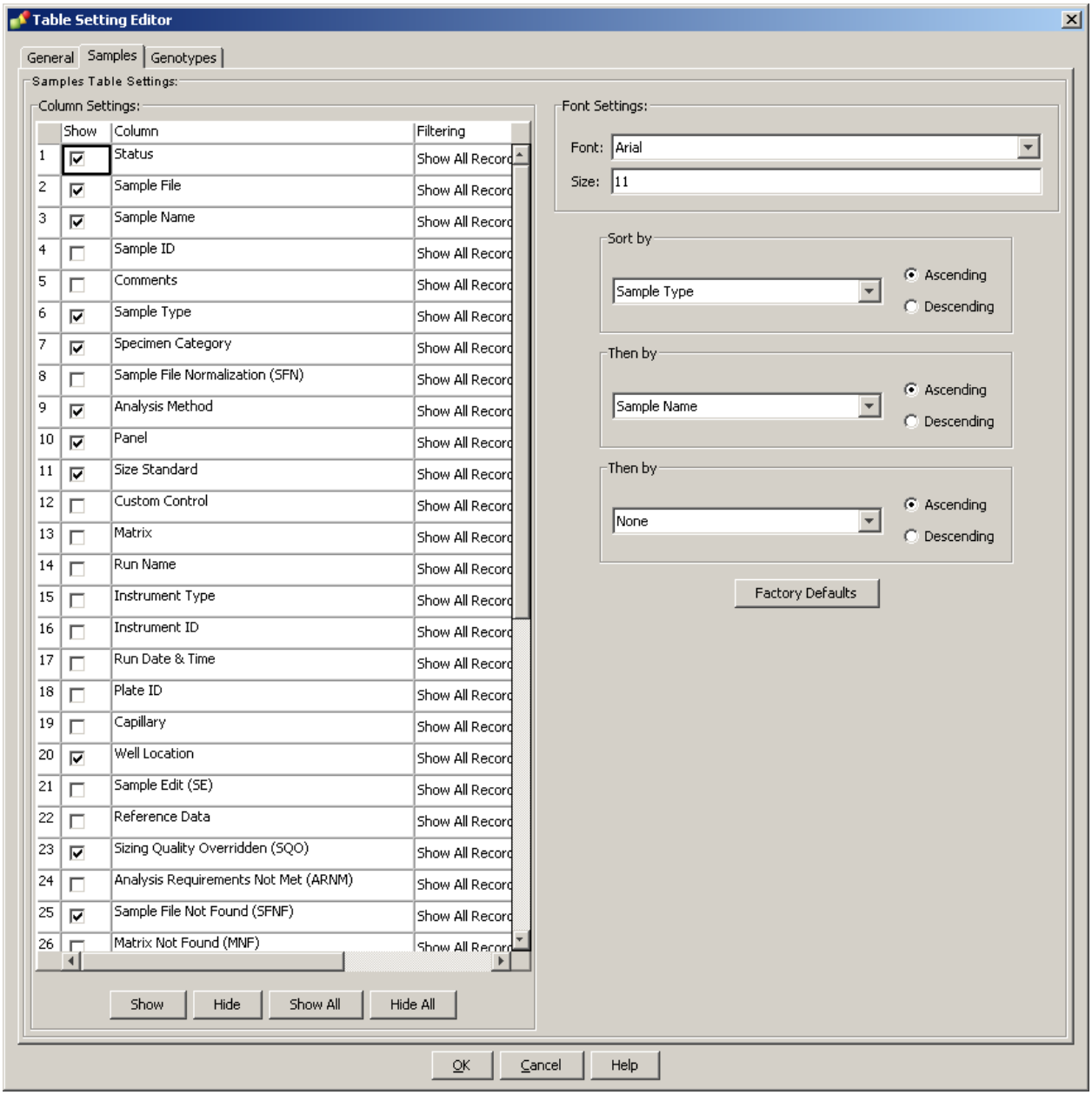
3.8.4.4.2 Select the “Table Setting” tab and click “New”.

3.8.4.4.3 Under the “General” tab name the Table Setting “CODIS”.

3.8.4.4.4 Select “ISP Databasing Security Group”.

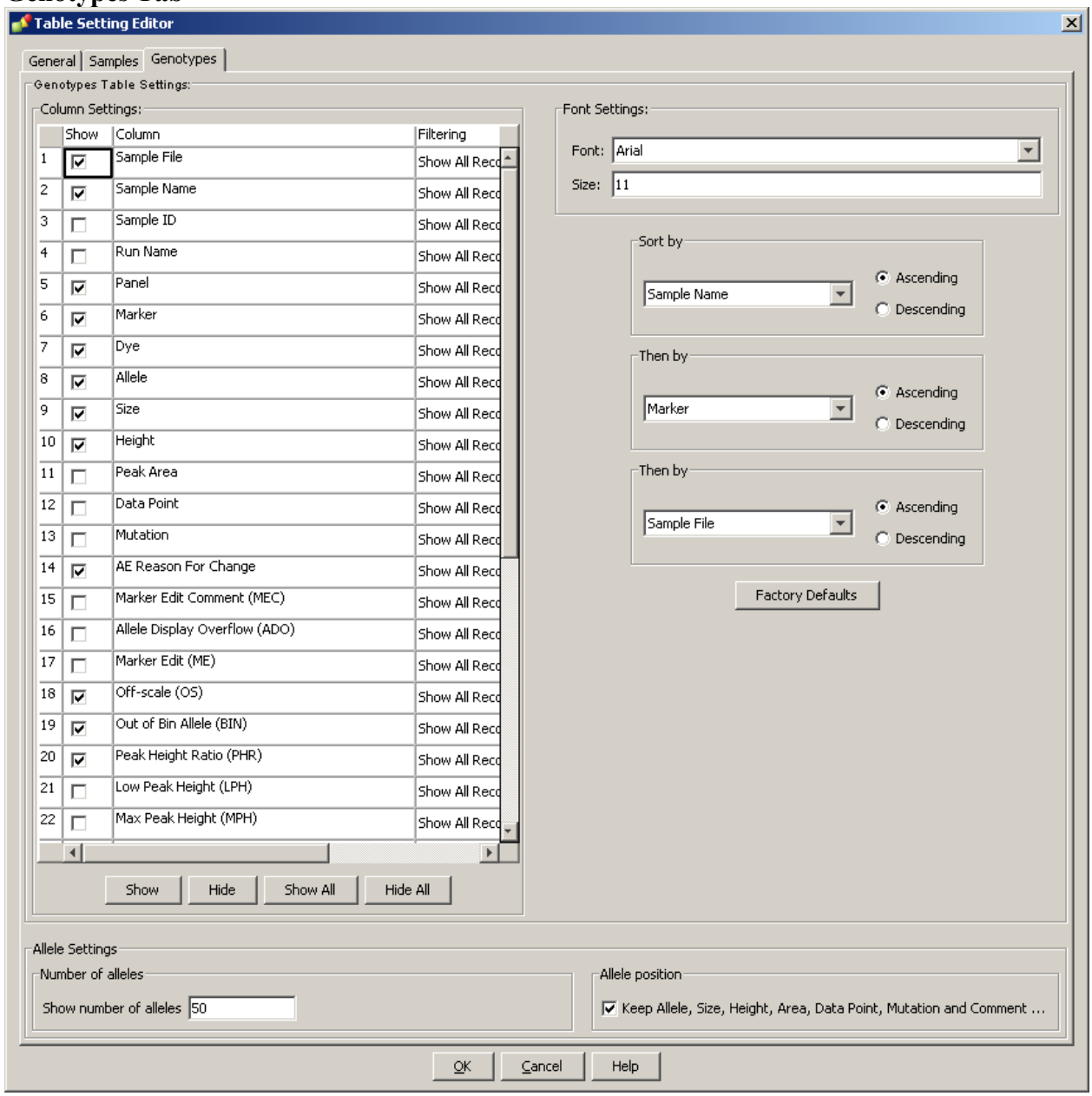
3.8.4.4.5 Select the “Samples” tab. Under column headings, ensure the following are checked: Status, Sample File, Sample Name, Sample Type, Specimen Category, Analysis Method, Panel, Size Standard, Well Location, Sample File Not Found, Sample Off-Scale, Sizing Quality, Composite GQ, User Defined Column 1 and User Defined Column 2. Change other settings to those shown below.

Samples Tab



3.8.4.4.6 Select the "Genotypes" tab. Under Column Settings, ensure the following are checked: Sample File, Sample Name, Panel, Marker, Dye, Allele Size, Height, AE Reason for Change, Off-Scale, Out of Bin Allele, Peak Height Ratio, Control Concordance and Genotype Quality. Change other settings to those shown below.

Genotypes Tab



3.8.4.5 Create a Plot Setting – Samples and Controls

3.8.4.5.1 Select “Tools”, then “GeneMapper Manager”.

3.8.4.5.2 Select the “Plot Setting” tab and click “New”.

3.8.4.5.3 Under the “General” tab, name the Plot Setting “CODIS”.

3.8.4.5.4 Select "ISP Databasing Security Group".

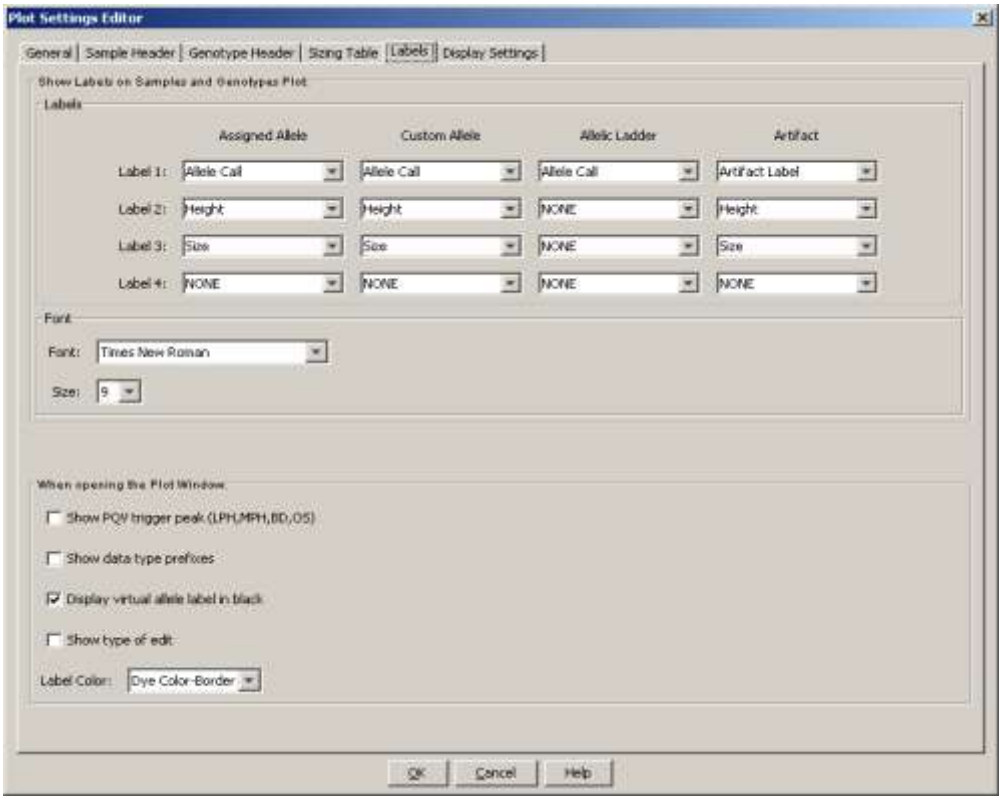
3.8.4.5.5 Select the "Sample Header" tab. Ensure the following are checked: Sample File, Sample Name, Panel, Sizing Quality Overridden, Off-Scale, Sizing Quality, Sample Spike, Mixed Source, Outside Marker Range and Composite GQ.

3.8.4.5.6 Select the "Genotype Header" tab. Ensure the following are checked: Sample File, Sample Name, Panel, Marker, Off-Scale, Out of Bin Allele, Peak Height Ratio, Low Peak Height, Allele Number, Broad Peak, Control Concordance and Genotype Quality.

3.8.4.5.7 Select the "Sizing Table" tab. Ensure the following are checked: Dye/Sample Peak, Sample File Name, Marker, Allele, Size, Height, Area and Data Point. Leave the font as Arial 11.

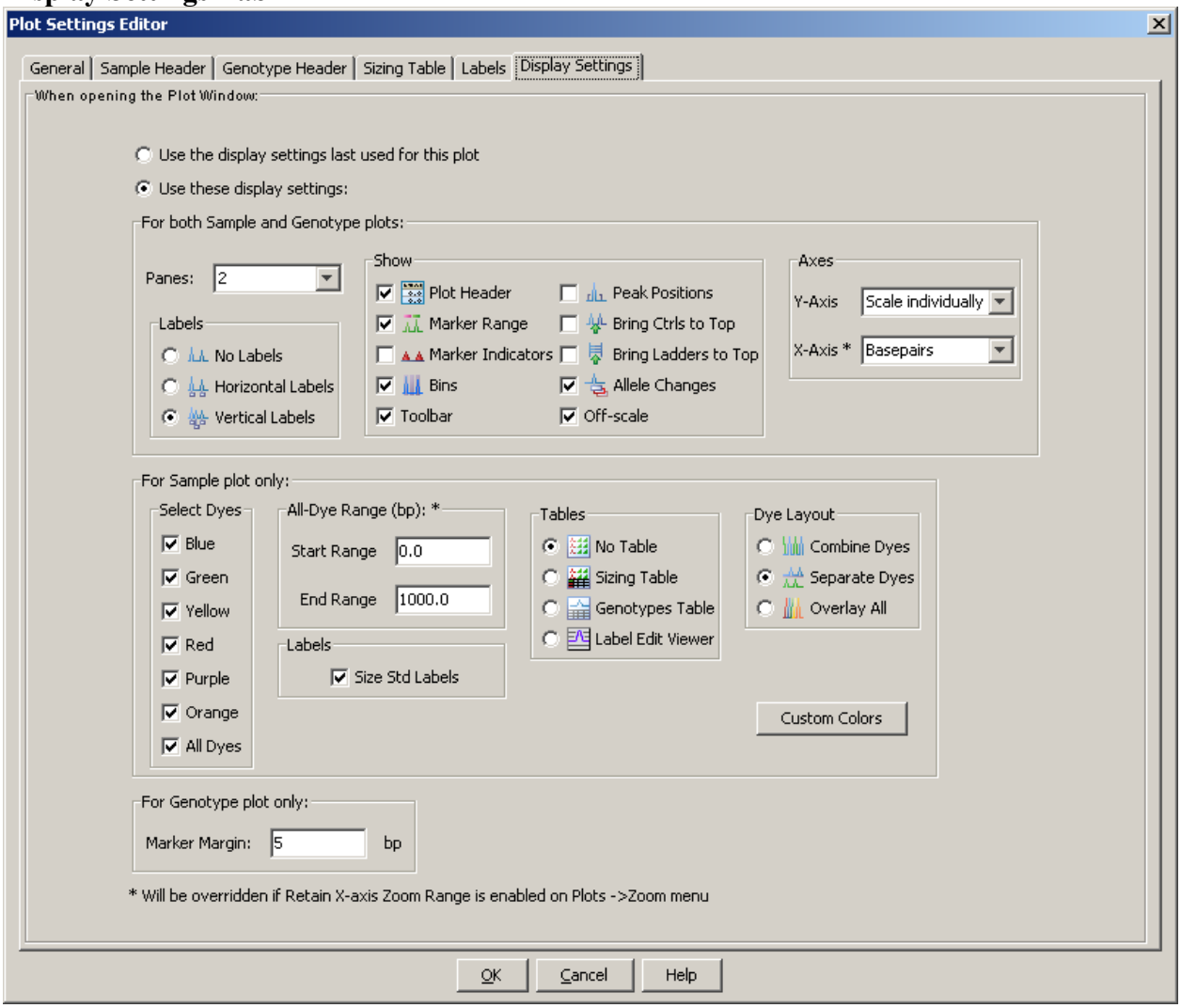
3.8.4.5.8 Select the "Labels" tab. Change the setting to those shown below.

Labels Tab



3.8.4.5.9 Select the "Display Settings" tab. Change the settings to those shown below.

Display Settings Tab



3.8.4.5.10 Select “OK”.

3.8.4.5.11 Click “DONE”.

3.8.5 Sampling

3.8.5.1 For oral swabs

3.8.5.1.1 Place the head of one swab in a deep well plate.

3.8.5.1.2 In each of two 50ml conicals, mix 50ml SEB + 250µl ProK.

3.8.5.1.3 Using a fresh pipette tip every time, place 1ml solution into each well containing a swab head.

3.8.5.1.4 For the reagent blank, place the solution only into well D12 for a full plate. A different well may be chosen for partial plates.

3.8.5.1.5 Incubate at 70° C for approximately 1 hour.

3.8.5.2 For stain cards

3.8.5.2.1 Using the 1.2mm Harris punch, take one punch of the stained area and place directly in the 96-well plate to be used in amplification set-up.

3.8.5.2.2 An unused stain card shall be punched between each stain card sample as a cleaning punch.

3.8.5.2.3 Repeat for all samples.

3.8.5.2.4 For the reagent blank, place one punch from an unused stain card in the appropriate well.

3.8.5.3 Wells A1, E12, F12, G12 and H12 of a full plate should be left empty for controls to be added later in the process.

3.8.6 Powerplex® 18D Amplification Set-Up

3.8.6.1 Full plates of oral swabs

3.8.6.1.1 The following steps shall be performed in the PCR amplification set-up area.

3.8.6.1.2 Thaw the Amplification Grade Water, PowerPlex® D 5X Master Mix and PowerPlex® 18D 5X Primer Pair Mix. When thawed, it is important to vortex the PowerPlex® D 5X Master Mix and PowerPlex® 18D 5X Primer Pair Mix tubes for 5 to 10 seconds. (Do not centrifuge the 5X

Master Mix or 5X Primer Pair Mix as this may cause the components to be concentrated at the bottom of the tube.) The Amplification Grade Water may be stored at 4° C for extended periods.

3.8.6.1.3 Place one 0.2ml 96-well plate in the PCR setup area and label appropriately.

3.8.6.1.4 To make a master mix, add the following to a tube and mix gently.

3.8.6.1.4.1 1300µl Amplification Grade Water

3.8.6.1.4.2 500µl PowerPlex® D 5X Master Mix

3.8.6.1.4.3 500µl PowerPlex® 18D 5X Primer Pair Mix

3.8.6.1.5 Add 23µl of PCR master mix to each well except A1 and H12 using a repeat pipettor.

3.8.6.1.6 Pipette 2µl of liquid from each sample into the respective well containing master mix, using a new tip for each sample.

3.8.6.1.7 For the positive control, dilute the 2800M DNA standard or other approved positive DNA standard supplied with the PowerPlex® 18D kit to 2.5ng/µl. Pipette 2µl into the designated well containing 23µl of PCR master mix. A positive control shall be included on each plate.

3.8.6.1.8 For the negative amplification control, pipette 2µl of nuclease free or amplification grade water into the designated well containing 23µl of the PCR master mix. A negative amplification control shall be included on each plate.

3.8.6.1.9 Place the 96-well plate in a thermal cycler.

3.8.6.1.10 Select and run the "codis-27 cycle" thermal cycling protocol.

3.8.6.1.11 Remove the plate after the amplification process is completed.

3.8.6.1.12 Store the amplified samples in the freezer or refrigerator (if they are to be used within 24 hours).

3.8.6.2 Partial plates or those containing a combination of oral and blood samples

3.8.6.2.1 The following steps shall be performed in the PCR amplification set-up area.

3.8.6.2.2 Thaw the Amplification Grade Water, PowerPlex® D 5X Master Mix and PowerPlex® 18D 5X Primer Pair Mix. When thawed, it is important to vortex the PowerPlex® D 5X Master Mix and PowerPlex® 18D 5X Primer Pair Mix tubes for 5 to 10 seconds. (Do not centrifuge the 5X Master Mix or 5X Primer Pair Mix as this may cause the components to be concentrated at the bottom of the tube.) The Amplification Grade Water may be stored at 4° C for extended periods.

3.8.6.2.3 Place one 0.2ml 96-well plate in the PCR setup area and label appropriately.

3.8.6.2.4 Determine the number of samples of each type to be amplified, including controls (reagent blank, positive control and amplification blank). Add 2 to 4 reactions to this number to compensate for the loss that occurs during reagent transfer.

3.8.6.2.5 Stain card punches require a separate master mix preparation from the master mix for oral swabs and controls.

3.8.6.2.6 For oral swabs and controls, calculate the required amount of each component of the PCR master mix. Multiply the volume (µl) per sample by the total number of reactions to obtain the final volume (µl).

3.8.6.2.6.1 13µl Amplification Grade Water

3.8.6.2.6.2 5µl PowerPlex® D 5X Master Mix

3.8.6.2.6.3 5µl PowerPlex® 18D 5X Primer Pair Mix

3.8.6.2.7 Add the calculated volume of each component to a tube. Mix gently.

3.8.6.2.8 Add 23µl of PCR master mix to each well that will contain sample using a repeat pipettor.

3.8.6.2.9 Pipette 2µl of liquid from each sample into the respective well containing master mix, using a new tip for each sample.

3.8.6.2.10 For the positive control, dilute the 2800M DNA standard or other approved positive DNA standard supplied with the PowerPlex® 18D kit to 2.5ng/µl. Pipette 2µl into the designated well containing 23µl of PCR master mix. A positive control shall be included on each plate.

3.8.6.2.11 For the negative amplification control, pipette 2µl of nuclease free or amplification grade water into the designated well containing 23µl of the PCR master mix. A negative amplification control shall be included on each plate.

3.8.6.2.12 For stain card punches, calculate the required amount of each component of the PCR master mix. Multiply the volume (µl) per sample by the total number of reactions to obtain the final volume (µl).

3.8.6.2.12.1 15µl Amplification Grade Water

3.8.6.2.12.2 5µl PowerPlex® D 5X Master Mix

3.8.6.2.12.3 5µl PowerPlex® 18D 5X Primer Pair Mix

3.8.6.2.13 Add the calculated volume of each component to a tube. Mix gently.

3.8.6.2.14 Add 25µl of PCR master mix to each well containing a stain card punch. Use a new disposable tip for each well.

3.8.6.2.15 Cover with foil seal and briefly spin down to remove air bubbles from the wells.

3.8.6.2.16 Place the 96-well plate in a thermal cycler.

3.8.6.2.17 Select and run the "codis-27 cycle" thermal cycling protocol.

3.8.6.2.18 Remove the plate after the amplification process is completed.

3.8.6.2.19 Store the amplified samples in the freezer or refrigerator (if they are to be used within 24 hours).

3.8.7 Applied Biosystems 3500xl Genetic Analyzer - Data Collection Software version 2.0 - PowerPlex® 18D Electrophoresis

3.8.7.1 Sample Preparation

3.8.7.1.1 **Note:** The quality of formamide is critical for the successful detection of a DNA profile. Deionized formamide shall be used that has a conductivity of less than 100µS/cm, such as Hi-Di™ Formamide. The formamide shall be frozen in aliquots at -20^{o}C and the remainder of each aliquot shall be discarded after it is thawed. Multiple freeze-thaw cycles or long-term storage at 4^{o}C may cause breakdown of the formamide

which can create ions that compete with DNA during injection. This will cause lower peak heights and decreased sensitivity.

3.8.7.1.2 **Caution:** Formamide is an irritant and teratogen; therefore universal precautions and a fume hood shall be utilized when manually working with formamide to avoid inhalation and contact with the skin.

3.8.7.1.3 Thaw the CC5 ILS-500, the allelic ladder and an aliquot of Hi-Di™ Formamide. When thawed, vortex to mix.

3.8.7.1.4 Prepare a loading cocktail by combining 100µl internal lane standard (CC5 ILS-500) with 1000µl Hi-Di™ Formamide.

3.8.7.1.4.1 If the plate is not full, determine the amount of ILS and formamide needed by the following formula: [(1µl ILS) x (# samples)] + [(10µl Hi-Di™ Formamide) x (# samples)]

3.8.7.1.5 Vortex to mix.

3.8.7.1.6 Pipette 11µl of the loading cocktail into each well.

3.8.7.1.7 Add 1µl of amplified sample or 1µl of the allelic ladder mix to each well, using a new tip for each sample. At least one allelic ladder is required within each run folder.

3.8.7.1.7.1 If the plate is not full, add formamide or loading cocktail into empty wells to complete an injection set of 24. Every well in which an injection is occurring must contain liquid.

3.8.7.1.7.2 It is recommended this step be witnessed if samples are being cherry picked from multiple amplifications.

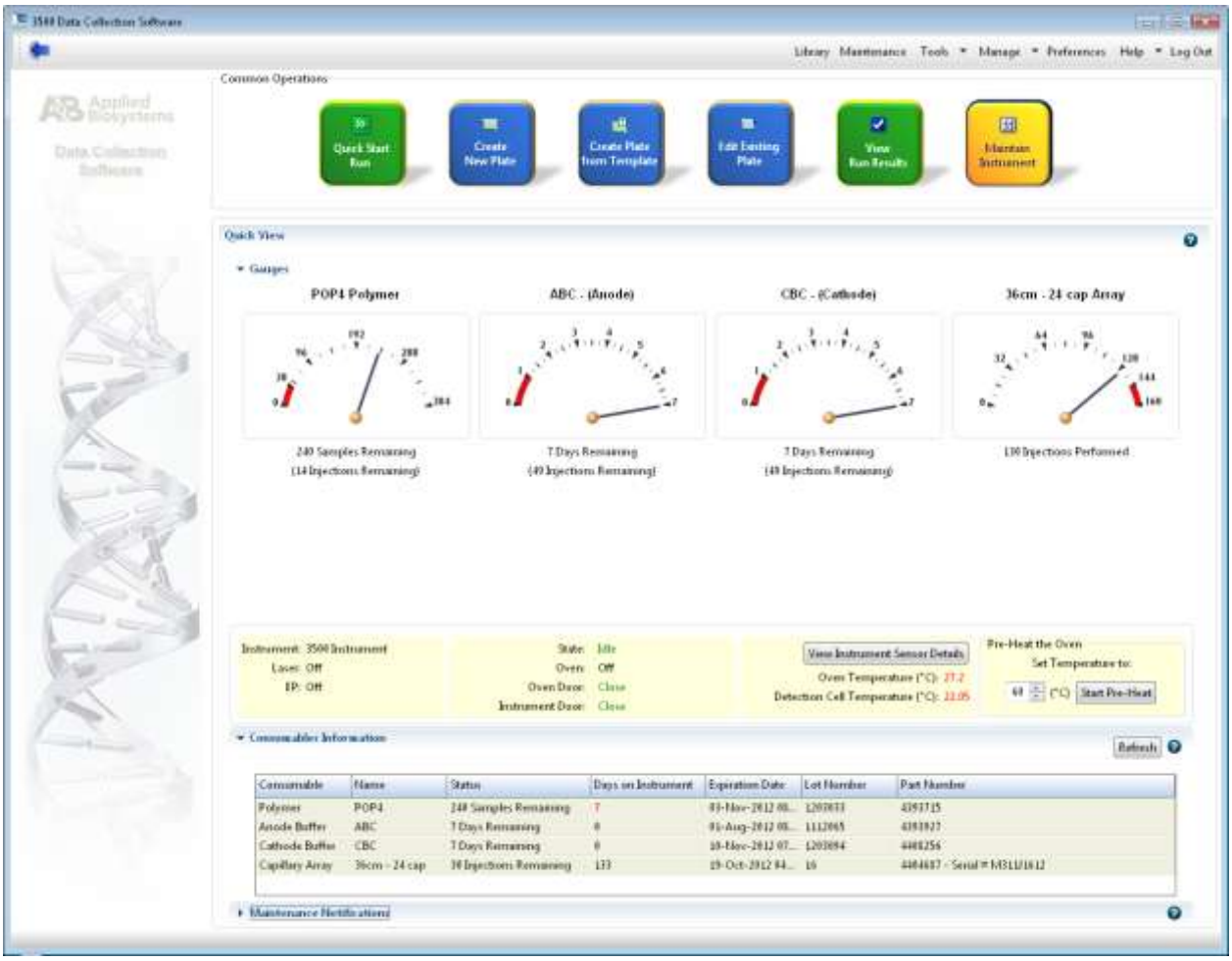
3.8.7.1.8 Cover the wells with the plate septa and briefly spin down to remove air bubbles from the wells.

3.8.7.1.9 Denature the samples at 95^{o}C for ~3 minutes, then immediately chill on crushed ice or a cold pack for ~3 minutes. Denature the samples just prior to loading the instrument. Avoid denaturing the samples for longer than 3 minutes as extended heat denaturing can lead to the appearance of artifacts.

3.8.7.2 Create a Plate

3.8.7.2.1 Open the 3500 Data Collection Software. The Dashboard screen will launch. Ensure that the Consumables Information and Maintenance Notifications are acceptable. The oven temperature should be set to 60°C.

Dashboard



3.8.7.2.2 Select “Start Pre-Heat”. This should be done at least 30 minutes prior to the first injection to preheat the oven.

3.8.7.2.3 Select “Create New Plate”. Alternatively, “Create Plate from Template” may be used.

3.8.7.2.3.1 Assign the plate name with the year, database batch number and run number (ex. 2012_DB001_01).

3.8.7.2.3.2 Select “96” for Number of Wells.

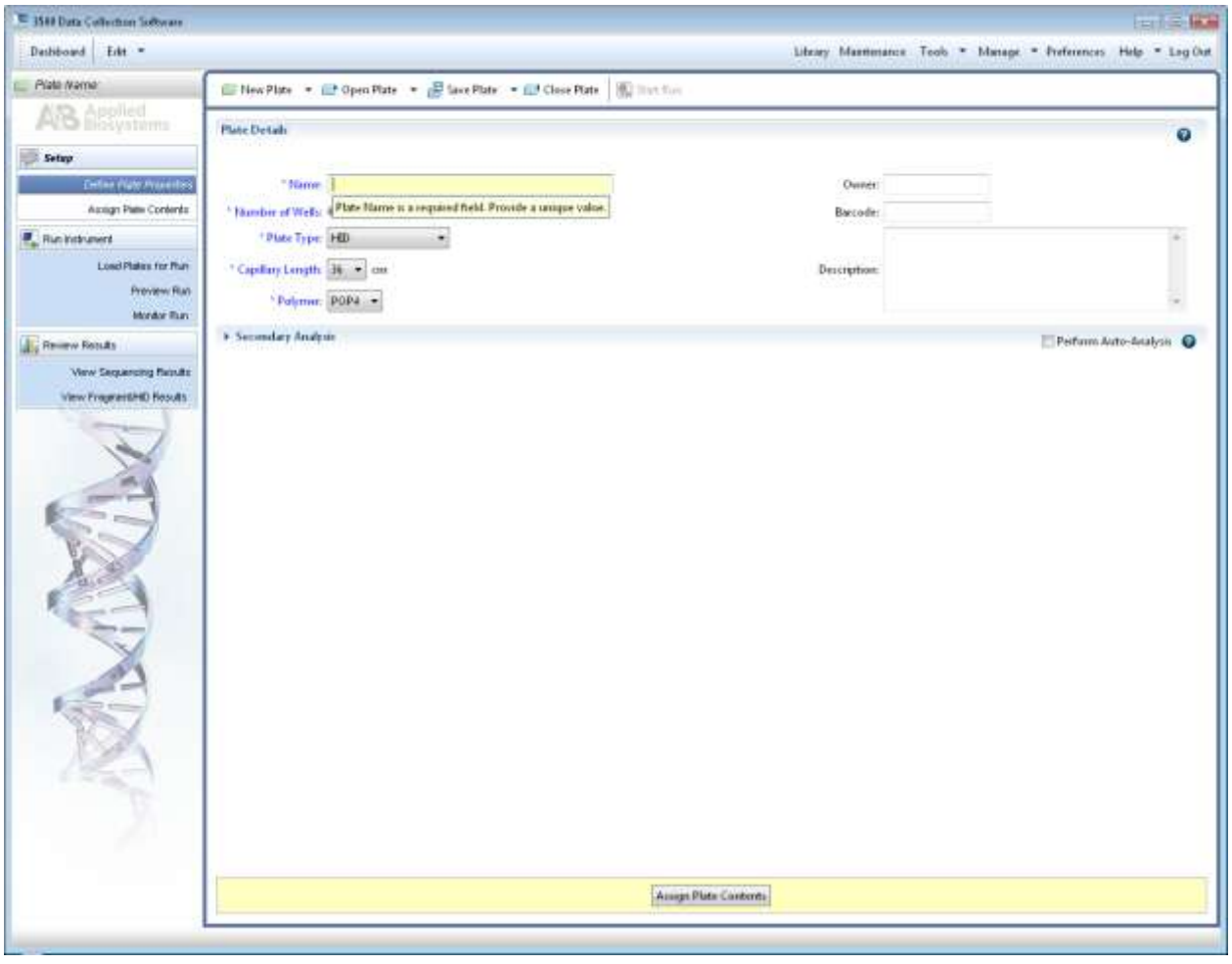
3.8.7.2.3.3 Select “HID” for Plate Type.

3.8.7.2.3.4 Select “36” for Capillary Length.

3.8.7.2.3.5 Select “POP4” for Polymer.

3.8.7.2.3.6 Type your initials in Owner box.

Create a Plate



3.8.7.2.4 Click on “Assign Plate Contents”.

3.8.7.2.5 Enter sample information or import a plate record.

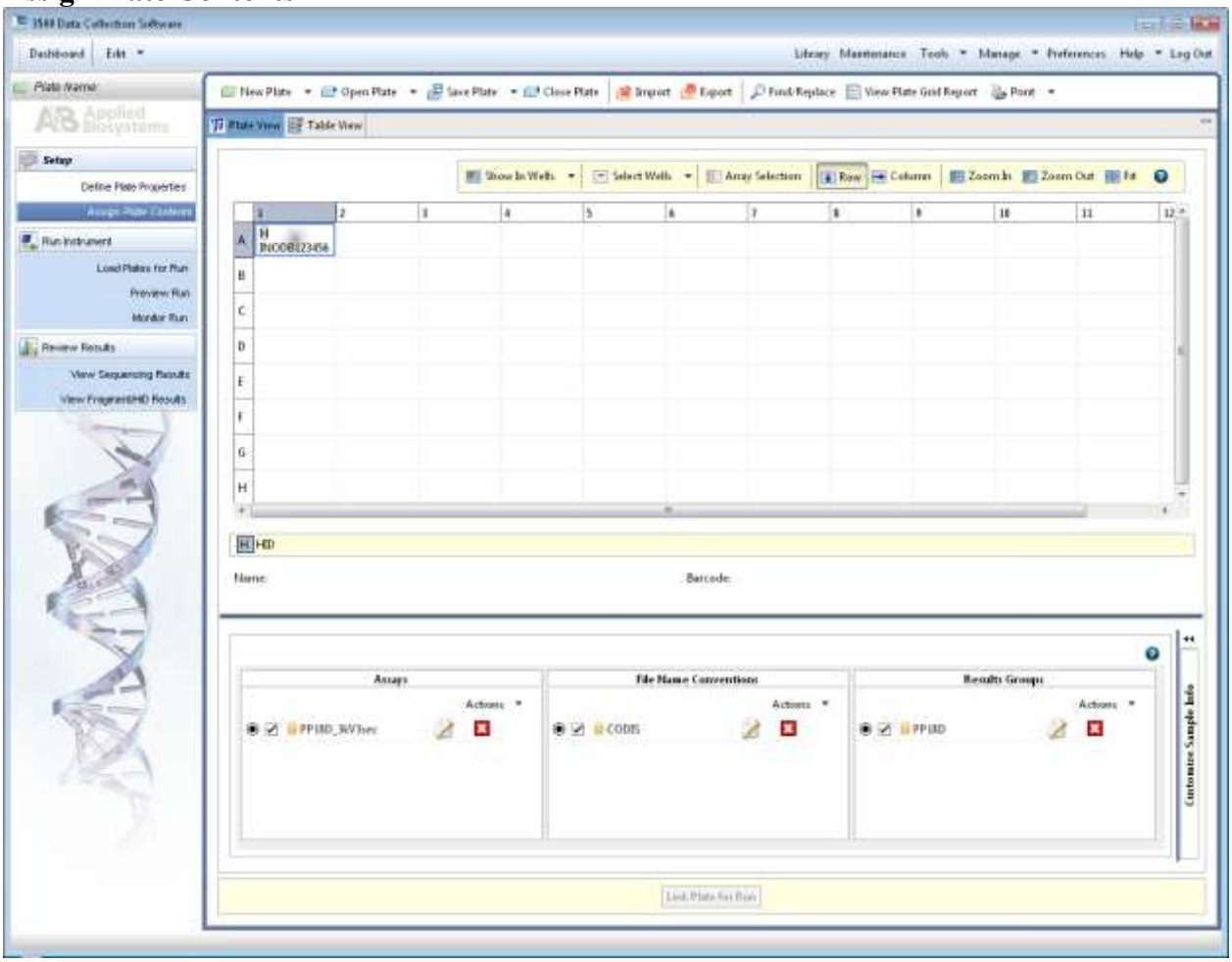
3.8.7.2.6 Under “Assays,” click “Add from Library” and select the appropriate assay. Click “Add to Plate” then “Close”.

3.8.7.2.6.1 To run different assays on different injections, click the "Actions" arrow to assign additional assays.

3.8.7.2.7 Under "File Name Convention", click "Add from Library" and select "CODIS". Click "Add to Plate" then "Close".

3.8.7.2.8 Under "Results Groups", click "Add from Library" and select "PP18D". Click "Add to Plate" then "Close".

Assign Plate Contents

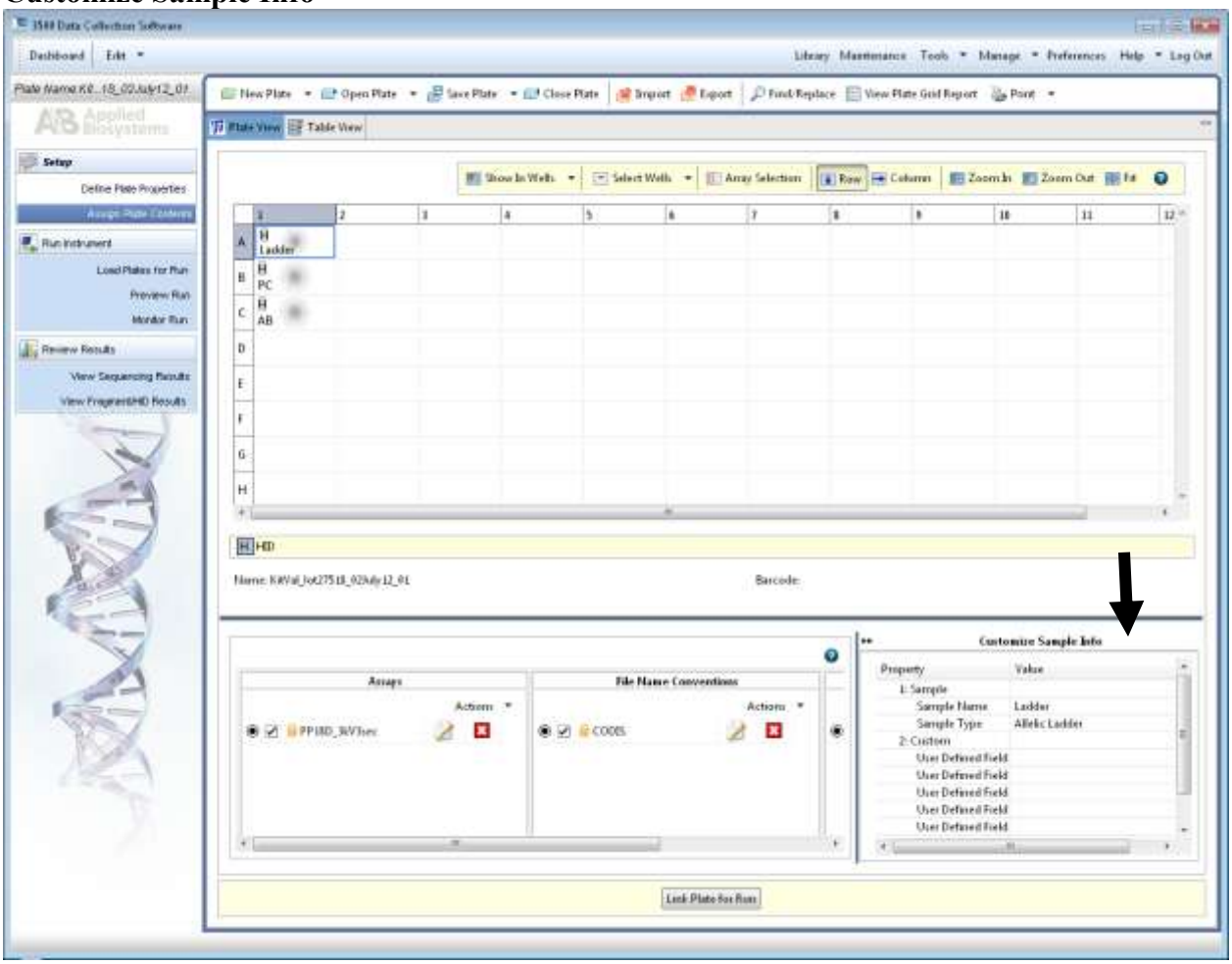


3.8.7.2.9 Expand "Customize Sample Info" window by clicking on the arrows in the lower right portion of the screen.

3.8.7.2.9.1 Use the drop-down box to select a sample type for each sample. For imported plates, this will be done automatically.

3.8.7.2.9.2 For all re-runs, the Batch# and Well# of the original location shall be typed into User Defined Field 2 (ex. 2012_DB001_A4).

Customize Sample Info



3.8.7.2.9.3 Minimize the Customized Samples window.

3.8.7.2.10 Highlight the sample wells, then select the boxes in the Assays, File Name Conventions and Results Groups that pertain to those samples.

3.8.7.2.11 Place plate on instrument in position A. Select “Link Plate for Run”. Click “OK”. The instrument automatically senses the plate and puts the information in the Plate A field. Click “OK”.

3.8.7.2.12 To add a second plate, follow steps 3.8.7.2.3 through 3.8.7.2.10. Place the plate in position B. Select the "Link Plate for Run" and click "OK". The instrument automatically senses the plate and puts the information in the Plate B field. Click "OK".

3.8.7.3 Start a Plate Run

3.8.7.3.1 A run name is automatically generated but can be modified if needed.

3.8.7.3.2 If you want to perform the injection in a certain order, click "Create Injection List" and use the arrows to re-order injections.

3.8.7.3.3 Click "Start Run".

3.8.7.3.4 After the run is complete, click unlink plate and remove it from the instrument. Plates may be stored in the freezer until no longer needed.

3.8.8 GeneMapper® *ID*-X Version 1.4 Software - PowerPlex® 18D Data Analysis

3.8.8.1 Processing Sample Data

3.8.8.1.1 Import the sample files from a single run folder by clicking on "Edit", then selecting "Add Samples to Project".

3.8.8.1.2 In the "Add Samples to Project" screen, navigate to the run folder that contains the sample files. If the entire run folder is to be imported, click on the folder to highlight it, then click the "Add to List" button at the bottom of the window. If only a portion of samples need to be selected, expand the folder to view the samples. Highlight the appropriate samples, ensuring that the allelic ladder, controls and all the desired samples are selected. Once all the samples are selected click the "Add to List" button at the bottom of the window.

3.8.8.1.3 There shall only be one injection parameter per project. A run folder shall not be created manually by manipulating sample files.

3.8.8.1.4 Ensure that the necessary files are now located in the "Samples to Add" window by double-clicking on the folder in the right pane, then click "Add".

3.8.8.1.5 After the samples have been added to the project, first briefly scan the raw data to ensure that a bad injection did not occur. To check the raw data, first expand the project folder in the left navigation pane, then click on a sample file, then click on the "Raw Data" tab in the right GeneMapper® *ID-X* window. To return to the "Samples" window, click on the project folder at the top of the left navigation pane.

3.8.8.1.6 The GeneMapper® *ID-X* project shall contain at least one allelic ladder from each run folder included in the project for proper genotyping. Multiple allelic ladders within a run folder will be averaged by the software to calculate the allelic bins. If a ladder injection is of low quality, delete the ladder or change the sample type from "Allelic Ladder" to "Sample" to remove it from consideration in calculating the bins.

3.8.8.1.7 Ensure the table setting at the top of the screen is set to "CODIS".

3.8.8.1.8 In the "Analysis Method" column, for each sample select "PP18D CODIS" from the drop-down menu. Click the column header cell to highlight the entire column, then select "Edit", then "Fill Down" (or the shortcut Ctrl + D).

3.8.8.1.9 In the "Panel" column, for each sample select "PowerPlex_18D_IDX_v1.2" from the drop-down menu. Click the column header cell to highlight the entire column, then select "Edit", then "Fill Down" (or the shortcut Ctrl + D).

3.8.8.1.10 In the "Size Standard" column, select "CC5_ILS500" from the drop-down menu. Click the column header cell to highlight the entire column, then select "Edit", then "Fill Down" (or the shortcut Ctrl + D).

3.8.8.1.11 The Analysis Method, Size Standard and Panel can be set as defaults when a GeneMapper® *ID-X* project is opened. Under the "File" menu, select "Project Options". Under the "Add Samples" tab select the above settings as the default in the drop-down menus for Analysis Method, Size Standard and Panel. Click "OK".

3.8.8.1.12 Select the green "Analyze" arrow button to start the data analysis. At the Project name prompt, save the project. At a minimum the project name shall contain the batch number and injection parameters for the project. Select the "ISP Databasing Security Group".

3.8.8.2 Evaluating Sample Data

3.8.8.2.1 The Sizing Quality PQV will be yellow or red if the Sizing Quality is less than 0.75. These samples should be interpreted with caution and may be re-run at the analyst's discretion.

3.8.8.2.2 Highlight all sample rows containing Allelic Ladders. Then click "Analysis", then "Display Plots". Magnify the area from about 100 bp to 500 bp. Check the allelic ladders to ensure that the correct allele calls are made for each peak. (Refer to the PowerPlex®18D System Technical Manual for current Allelic Ladder allele calls.) Close out of the Samples Plot window.

3.8.8.2.3 Highlight all sample rows containing negative controls (ex. amplification blanks and reagent blanks). Then click "Analysis", then "Display Plots". Check the negative controls to ensure that no peaks above threshold are present. Close out of the Samples Plot window.

3.8.8.2.4 Highlight all sample rows containing positive controls. Then click "Analysis", then "Display Plots". Magnify the area from approximately 100 bp to 500 bp. Check the positive controls to ensure that all allele calls are correct. At least one positive control sample shall pass for each amplification. Close out of the Samples Plot window.

3.8.8.2.5 Highlight all remaining sample rows. Then click "Analysis", then "Display Plots". Magnify the area from approximately 100 bp to 500 bp. Evaluate all allele calls.

3.8.8.2.6 After all analysis is complete, save the Run Folder containing the sample files and associated projects under the batch number in the zCODIS folder located on the server. Periodically delete projects from GeneMapper® ID-X Manager to maintain computer disk space. Data files will be deleted from the database on a routine basis.

3.8.8.2.7 Create a CMF for import into CODIS.

3.8.8.2.7.1 Click on "Tools" then "CODIS Export Manager".

3.8.8.2.7.2 In both the "Source Lab IDs" and "Destination Lab IDs" windows, enter "INISP5200".

3.8.8.2.7.3 Click "OK".

3.8.8.2.7.4 Set Specimen Category for all passing offender samples as "Convicted Offender". Specimen Category for samples to be re-

run and controls shall be "No Export" to exclude them from the CMF.

3.8.8.2.7.5 Click "File" then "Export Table for CODIS".

3.8.8.2.7.6 Choose Export File As "CMF 3.2" and both Source and Destination Lab as "INISP5200".

3.8.8.2.7.7 After import, the CMF shall be saved to the batch file on the server. Name the CMF with, at minimum the batch number and injection protocol (ex. 2012_DB001_3kV3sec). The plate injection number should also be included in cases where more than one CMF is created for a single plate.

3.8.9 Interpretation Guidelines for PowerPlex® 18D

3.8.9.1 Once a determination has been made whether a peak is to be considered a true allele, the following interpretation guidelines shall be used. The minimum peak height threshold is established at 100 relative fluorescent units (RFU) for GeneMapper® *ID-X* software. The analytical threshold for data interpretation is 100 RFU. The stochastic threshold for data interpretation is 250 RFU.

3.8.9.2 Peaks below 100 RFU shall not be interpreted. If allelic drop-out is suspected in any of the CODIS core loci, the sample shall be re-run when possible to attempt to obtain a full profile. If allelic drop-out is suspected in a non-core locus, that locus may be omitted from CODIS entry and allele calls deleted.

3.8.9.3 Samples with a single peak between 100 RFU and 250 RFU should be interpreted with caution and may be re-run at the analyst's discretion. If the single peak in this range is in a non-core locus, that locus may be omitted from CODIS entry and allele call deleted.

3.8.9.4 Controls

3.8.9.4.1 Failed controls require notification to the Technical Leader with appropriate documentation in the batch folder located on the server.

3.8.9.4.2 The appearance of pull-up or known artifact peaks does not render the following controls inconclusive.

3.8.9.4.3 **Reagent Blank**

3.8.9.4.3.1 The purpose of the reagent blank is to determine if the reagents used to extract the associated samples were contaminated by human DNA. Therefore no signal should be detected in this sample well other than the internal lane standard. If a signal is detected in the reagent blank, all results of samples associated with that reagent blank shall be considered inconclusive.

3.8.9.4.3.2 A reagent blank with peaks below 100 RFU shall not prevent associated samples from being interpreted.

3.8.9.4.3.3 A reagent blank with peaks of 100 RFU and above shall be considered a failed negative control. All associated samples shall be inconclusive. All the samples shall be repeated when appropriate.

3.8.9.4.4 **Positive Control**

3.8.9.4.4.1 The 2800M positive DNA control supplied with the PowerPlex® 18D kits is used as a positive control to demonstrate that the kit is performing properly. If the expected alleles (see table below) are not detected in at least one positive control well, then the test is considered inconclusive.

STR Locus	2800M
D3S1358	17,18
TH01	6,9.3
D21S11	29,31.2
D18S51	16,18
Penta E	7,14
D5S818	12,12
D13S317	9,11
D7S820	8,11
D16S539	9,13
CSF1PO	12,12
Penta D	12,13
Amelogenin	XY
vWA	16,19
D8S1179	14,15
TPOX	11,11
FGA	20,23
D19S433	13, 14
D2S1338	22, 25

3.8.9.4.5 **Amplification Blank**

3.8.9.4.5.1 The purpose of the amplification blank is to determine if human DNA contaminated the samples at the amplification step. Because no template DNA was placed in the reaction, the sample well should be blank except for the internal lane standard peaks. If amplified product is detected in the amplification blank well, the test is considered inconclusive.

3.8.9.4.5.2 An amplification blank with peaks below 100 RFU shall not prevent associated samples from being interpreted.

3.8.9.4.5.3 An amplification blank with peaks of 100 RFU and above shall be considered a failed negative control. All associated samples shall be inconclusive. All the samples shall be repeated when appropriate.

3.8.9.5 The analytical and stochastic threshold shall be determined during validation. It shall be at the analyst's discretion, based on experience and training, as to which peaks are suitable for interpretation. If an analyst has determined that a peak that has been labeled by the GeneMapper *ID-X* software is not a true allele peak, the analyst can delete the allele call label in the software. The GeneMapper *ID-X* software is set to record all allele changes. Therefore, any change or deletion in an allele call shall be maintained in the saved project.

3.8.9.6 An analyst is required to visually confirm all allelic ladders used for allele designation performed correctly and the allele calls for all positive controls are correct.

3.8.9.7 **A Global filter** shall be set in the GeneMapper® *ID-X* software to 20%. Therefore, any peaks that are less than 20% of the major peak at a locus will be automatically filtered out by the software and not labeled. This should significantly reduce the frequency of many of the following types of peaks that will be labeled as alleles by the software.

3.8.9.8 **Stutter peaks** are artifacts of the amplification process. These peaks will typically be observed in the n-4 position of major peaks for tetranucleotide repeat loci or in the n-5 position of major peaks for the pentanucleotide repeat loci. The peak heights of stutter peaks will be less intense than that of the major peak. It is the analyst's discretion to determine which allele calls may be deleted in GeneMapper® *ID-X* analysis.

3.8.9.8.1 **Stutter peaks** have also been documented at the n+4 or n+5 position. These peaks will also have significantly less intense signal than the major peak. Other artifacts of less intensity have been reported which may not

line up with the ladder. The interpretation of these peaks, similar to the other artifact peaks, shall be at the discretion of the analyst based on their training and experience.

3.8.9.9 **Artifacts** have been observed and documented utilizing the PowerPlex® 18D amplification kit. The intensity of these peaks is directly related to signal intensity; therefore, reducing the signal intensity below 10000 RFU should minimize the appearance of these types of artifacts.It is the analyst's discretion to determine which allele calls may be deleted in GeneMapper® *ID-X* analysis. Examples of documented artifacts can be found in the PowerPlex® 18D System Technical Manual.

3.8.9.10 **Pull-up or bleed through** peaks can occur if signal intensity of sample or ILS peaks is too high or if a new spectral calibration needs to be run. Any pull-up peaks called as alleles by the GeneMapper® *ID-X* software should be deleted . The sample should be re-run if a pull-up peak interferes with the analyst's ability to evaluate the profile based on their experience and training.

3.8.9.11 **Spikes** are peaks that generally appear in all colors and are sharper than regular peaks; however, they can occur predominantly in one color. Spikes are a natural consequence of capillary electrophoresis and can be caused by dust present in the system as well as urea crystals in the system. It is essential that the instrumentation be maintained and cleaned regularly to minimize the appearance of spikes. All spikes called as alleles by the GeneMapper® *ID-X* software should be deleted . A sample should be re-injected when a spike interferes with the analyst's ability to evaluate the profile based on their experience and training.

3.8.9.12 **Rare variants** (microvariants) have been described in the literature. These peaks will have a similar intensity to the other major peak for that locus but will not line up with the allelic ladder or have a bin in the GeneMapper® *ID-X* PowerPlex® 18D panels and bins settings.

3.8.9.12.1 Alleles one, two or three nucleotides shorter than the common four base repeat alleles (or four nucleotides shorter in the case of five base repeat alleles) which are located between two alleles on the ladder shall be described as the short repeat followed by the number of base pairs it is larger (a 0.1, 0.2, 0.3, or 0.4 in the case of a pentanucleotide repeat). Therefore, if a peak is 1 base pair larger than the 5 allele it shall be designated as 5.1. The precision of sizing at a 99.7% confidence level is less than 0.25 bp which is precise enough to be confident in the sizing of microvariants. A microvariant 4 base pairs larger than an allele (or 5 base pairs for a pentanucleotide) on the ladder may be designated with the full

repeat number (A peak 4 base pairs larger than the 5 allele could be designated a 6; 5 base pairs larger a 6.1).

3.8.9.12.2 Alleles which are located outside the range of the ladder or bin set (above or below) shall be designated as "<" or ">" the smallest or largest allele for that locus.

3.8.9.12.3 Any allele peak that is not present in the allelic ladder and does not have an associated "bin" in the GeneMapper® *ID-X* analysis software, will be called "OL" by the software. The analyst can rename the allele.

3.8.9.13 Mixed DNA Samples shall be investigated for possible contamination and re-run as appropriate. All loci shall be taken into consideration when determining if a mixture is present. A three-peak pattern at one or more loci may be an indication of a mixture. However, three-peak patterns have been reported for single-source stains, but these instances are rare. If a three-peak pattern is observed for a single locus and is believed to be tri-allelic, the profile should be re-amplified and re-analyzed to confirm the possible tri-allelic locus.

3.8.9.14 If it is determined a sample needs to be re-run, it shall be labeled "No Export" in the GeneMapper® *ID-X* analysis software and entered on the current re-analysis plate record(s).

3.8.10 General Rules for PowerPlex® 18D Analysis On The Applied Biosystems 3500xl Genetic Analyzer

3.8.10.1 If a sample is to be re-injected at higher injection parameters, the reagent blank and the amplification blank associated with that sample shall also be re-injected at the higher parameters. The positive control need not be injected at the same parameters as the samples associated with it.

3.8.10.2 If a selection of samples from an amplification requires re-injection at higher injection parameters with their associated blanks (as above) and the blanks at the higher parameters demonstrate contamination, the Technical Leader shall be informed. Sample data from the higher injection parameter shall be declared inconclusive. Notification to the Technical Leader shall be documented in the batch folder located on the server.

3.9 Records

3.9.1 The appropriate worksheets as contained in the Worksheet Manual or the equivalent workbooks shall be used to record all procedures.

3.9.2 The technical review of data shall be recorded on the worksheet provided for that purpose.

3.9.3 All data (3500xl run folders, GeneMapper® *ID-X* project files and CMFs) and documentation (plate records, emails, technical review worksheets) shall be saved in each batch folder located in the zCODIS folder on the server. All files stored on the server shall be routinely backed up to ensure security of data.

3.10 Interpretations of Results

3.10.1 Profiles with conclusive results at all CODIS core loci shall be entered into SDIS.

3.10.2 If test results from a sample cannot be clearly interpreted, that sample should be repeated. An additional collection from the individual may be requested.

3.10.3 See Interpretation guidelines in 3.8.9 and 3.8.10 for specific guidelines.

3.11 Report Writing: None

3.12 References

3.12.1 Federal Bureau of Investigation. Procedures for the Detection of Restriction Fragment Length Polymorphisms in Human DNA. FBI Laboratory. 1990.

3.12.2 Federal Bureau of Investigation. PCR-Based Typing Protocols. FBI Laboratory. 1994.

3.12.3 Promega Corporation. PowerPlex®18D System Technical Manual. Part No. TMD 031. Most current issue.

3.12.4 Promega Corporation. PowerPlex® 5-dye Matrix Standards, 3100/3130 Technical Bulletin. Part No. TBD024. Most current issue.

3.12.5 Applied Biosystems. 3500/3500xl Genetic Analyzer User Guide. 2010 or most current version.

3.12.6 Applied Biosystems. GeneMapper® *ID-X* Software Version 1.2 Reference Guide. Part Number 4426481, Rev. A. Foster City, CA. 10/2009.

3.12.7 Applied Biosystems. GeneMapper® ID-X Software Version 1.4 User Bulletin. Part No. 77684 Rev. A. Foster City, CA. 08/2012.

3.12.8 Bär W. et al. DNA recommendations: Further report of the DNA Commission of the ISFH regarding the use of short tandem repeat systems. *Int. J. Legal Med.* (1997) 110, 175-176.

3.12.9 Gill P. et al. Considerations from the European DNA profiling group (EDNAP) concerning STR nomenclature. *Forensic Science International* (1997) 87, 185-192.

3.12.10 Oostdik, K., Ensenberger, M., Krenke, B., Sprecher, C. and Storts, D. "The PowerPlex® 18D System: A Direct Amplification STR System with Reduced Thermal Cycling Time." *Profiles in DNA,* 2011, Promega Corporation.

Appendix 1

IC 10-13-6-1
"Combined DNA Index System"

Sec. 1. As used in this chapter, "Combined DNA Index System" refers to the Federal Bureau of Investigation's national DNA identification index system that allows the storage and exchange of DNA records submitted by state and local forensic DNA laboratories.
As added by P.L.2-2003, SEC.4.

IC 10-13-6-2
"DNA"

Sec. 2. As used in this chapter, "DNA" means deoxyribonucleic acid that:

(1) is located in the nucleated cells;

(2) provides an individual's personal genetic blueprint; and

(3) encodes genetic information that is the basis of human heredity and forensic identification.
As added by P.L.2-2003, SEC.4.

IC 10-13-6-3
"DNA analysis"

Sec. 3. As used in this chapter, "DNA analysis" means an identification process in which the unique genetic code of an individual that is carried by the individual's DNA is compared with the genetic codes of another individual.
As added by P.L.2-2003, SEC.4.

IC 10-13-6-4
"DNA profile"

Sec. 4. As used in this chapter, "DNA profile" means the results of all DNA identification tests on an individual's DNA sample.
As added by P.L.2-2003, SEC.4.

IC 10-13-6-5
"DNA record"

Sec. 5. As used in this chapter, "DNA record" refers to DNA identification information stored in the state DNA data base or the Combined DNA Index System for the purpose of generating investigative leads or supporting statistical interpretation of DNA test results that:

(1) is the result obtained from DNA typing tests; and

(2) is comprised of the characteristics of a DNA sample that are of value in

establishing the identity of individuals.
As added by P.L.2-2003, SEC.4.

IC 10-13-6-6
"DNA sample"

Sec. 6. As used in this chapter, "DNA sample" means a blood, tissue, or other body fluid sample:

(1) provided by a person with respect to offenses covered by this chapter; or

(2) submitted to the state police laboratory under this chapter for analysis or storage, or both.

As added by P.L.2-2003, SEC.4.

IC 10-13-6-7
"Superintendent"

Sec. 7. As used in this chapter, "superintendent" includes the superintendent or the superintendent's designee.
As added by P.L.2-2003, SEC.4.

IC 10-13-6-8
Establishment of DNA data base; mandatory and discretionary testing and analysis

Sec. 8. (a) The superintendent may establish a data base of DNA identification records of:

(1) convicted criminals;

(2) crime scene specimens;

(3) unidentified missing persons; and

(4) close biological relatives of missing persons.

(b) The superintendent shall maintain the Indiana DNA data base.

(c) The superintendent may contract for services to perform DNA analysis of convicted offenders under section 10 of this chapter to assist federal, state, and local criminal justice and law enforcement agencies in the putative identification, detection, or exclusion of individuals who are subjects of an investigation or prosecution of a sex offense, a violent crime, or another crime in which biological evidence is recovered from the crime scene.

(d) The superintendent:

(1) may perform or contract for performance of testing, typing, or analysis of a DNA sample collected from a person described in section 10 of this chapter at any time; and

(2) shall perform or contract for the performance of testing, typing, or analysis of a DNA sample collected from a person described in section 10 of this

chapter if federal funds become available for the performance of DNA testing, typing, or analysis.

(e) The superintendent shall adopt rules under IC 4-22-2 necessary to administer and enforce the provisions and intent of this chapter.

(f) The detention, arrest, or conviction of a person based on a data base match or data base information is not invalidated if a court determines that the DNA sample was obtained or placed in the Indiana DNA data base by mistake.

As added by P.L.2-2003, SEC.4. Amended by P.L.69-2005, SEC.1 and P.L.142-2005, SEC.1.

IC 10-13-6-9
Duties of superintendent

Sec. 9. The superintendent shall ensure that the Indiana DNA data base:

(1) supports development of a population statistics data base when personal identifying information is removed;

(2) supports identification research and protocol development of forensic DNA analysis;

(3) assists in achieving quality control; and

(4) assists in the recovery or identification of human remains from mass disasters or for other humanitarian purposes, including identification of missing persons who may be alive.

As added by P.L.2-2003, SEC.4.

IC 10-13-6-9.5
DNA sample processing fund

Sec. 9.5. (a) The DNA sample processing fund is established for the purpose of funding the collection, shipment, analysis, and preservation of DNA samples and the conduct of a DNA data base program under this chapter. The fund shall be administered by the superintendent.

(b) The expenses of administering the fund shall be paid from money in the fund.

(c) The treasurer of state shall invest the money in the fund not currently needed to meet the obligations of the fund in the same manner as other public money may be invested.

(d) Money in the fund at the end of a state fiscal year does not revert to the state general fund.

As added by P.L.176-2005, SEC.1.

IC 10-13-6-10
Persons required to provide DNA sample

Sec. 10. (a) This section applies to the following:

(1) A person convicted of a felony under IC 35-42 (offenses against the person) or IC 35-43-2-1 (burglary):

(A) after June 30, 1996, whether or not the person is sentenced to a term of imprisonment; or

(B) before July 1, 1996, if the person is held in jail or prison on or after July 1, 1996.

(2) A person convicted of a criminal law in effect before October 1, 1977, that penalized an act substantially similar to a felony described in IC 35-42 or IC 35-43-2-1 or that would have been an included offense of a felony described in IC 35-42 or IC 35-43-2-1 if the felony had been in effect:

(A) after June 30, 1998, whether or not the person is sentenced to a term of imprisonment; or

(B) before July 1, 1998, if the person is held in jail or prison on or after July 1, 1998.

(3) A person convicted of a felony, conspiracy to commit a felony, or attempt to commit a felony:

(A) after June 30, 2005, whether or not the person is sentenced to a term of imprisonment; or

(B) before July 1, 2005, if the person is held in jail or prison on or after July 1, 2005.

(b) A person described in subsection (a) shall provide a DNA sample to the:

(1) department of correction or the designee of the department of correction if the offender is committed to the department of correction; or

(2) county sheriff or the designee of the county sheriff if the offender is held in a county jail or other county penal facility, placed in a community corrections program (as defined in IC 35-38-2.6-2), or placed on probation.

A person is not required to submit a blood sample if doing so would present a substantial and an unreasonable risk to the person's health.

(c) The detention, arrest, or conviction of a person based on a data base match or data base information is not invalidated if a court determines that the DNA sample was obtained or placed in the Indiana DNA data base by mistake.

As added by P.L.2-2003, SEC.4. Amended by P.L.69-2005, SEC.2 and P.L.142-2005, SEC.2.

IC 10-13-6-11

Guidelines for DNA sample collection and shipment

Sec. 11. (a) The superintendent may issue specific guidelines relating to

procedures for DNA sample collection and shipment within Indiana for DNA identification testing.

(b) The superintendent shall issue specific guidelines related to procedures for DNA sample collection and shipment by the county sheriff or designee of the county sheriff under section 10(b)(2) of this chapter. The superintendent shall provide each county sheriff with the guidelines issued under this subsection. A county sheriff shall collect and ship DNA samples in compliance with the guidelines issued under this subsection.

(c) The superintendent may delay the implementation of the collection of DNA samples under section 10(b)(2) of this chapter in one (1) or more counties until the earlier of the following:

(1) A date set by the superintendent.

(2) The date funding becomes available by grant through the criminal justice institute.

If the superintendent delays implementation of section 10(b)(2) of this chapter or terminates a delay under section 10(b)(2) of this chapter in any county, the superintendent shall notify the county sheriff in writing of the superintendent's action.

As added by P.L.2-2003, SEC.4.

IC 10-13-6-12

Collection of samples

Sec. 12. DNA samples for the Indiana DNA data base must be collected in a medically approved manner by one (1) of the

following:

(1) A physician.

(2) A registered nurse.

(3) A licensed vocational nurse.

(4) A licensed clinical laboratory technologist.

(5) Any other person trained to collect DNA samples properly.

As added by P.L.2-2003, SEC.4.

IC 10-13-6-13

Purposes of testing

Sec. 13. (a) Tests performed on the DNA samples are for the following purposes:

(1) To analyze and type the genetic markers contained in or derived from DNA.

(2) For law enforcement identification purposes.

(3) For research or administrative purposes, including:

(A) development of a population statistics data base after personal

identifying information is removed;

(B) support of identification research and protocol development of forensic DNA analysis methods;

(C) quality control; and

(D) assisting in the recovery or identification of human remains from mass disasters or for other humanitarian purposes, including identification of missing persons who may be alive.

(b) Tests performed under this chapter must be conducted in a manner that produces compatible results with procedures specified by the Federal Bureau of Investigation Laboratory to ensure that DNA records are fully exchangeable between DNA laboratories.

As added by P.L.2-2003, SEC.4.

IC 10-13-6-14

Adherence to nationally recognized standards

Sec. 14. (a) A laboratory conducting forensic DNA analysis in Indiana must implement and follow nationally recognized standards for DNA quality assurance and proficiency testing, such as those approved by the American Society of Crime Laboratory Directors Laboratory Accreditation Board.

(b) Quality assurance guidelines issued by the Technical Working Group on DNA Analysis Methods serve as the standard for DNA testing under this chapter until national standards are set.

(c) A laboratory conducting forensic DNA analysis in Indiana shall forward relevant DNA data base records to the state police laboratory.

As added by P.L.2-2003, SEC.4.

IC 10-13-6-15

Disclosure of DNA samples and analysis

Sec. 15. A laboratory conducting forensic DNA analysis in Indiana may disclose or allow access to collected DNA samples and

DNA analysis results only under the following circumstances:

(1) To criminal justice agencies for law enforcement identification purposes.

(2) To defense counsel for criminal defense purposes.

(3) Upon authorization by a court or statute.

(4) For a population statistics data base, identification research and protocol development, or quality control purposes, but only if personal identifying information is removed.

(5) For purposes of postconviction DNA testing and analysis under IC 35-38-7.

As added by P.L.2-2003, SEC.4.

IC 10-13-6-16

Collection of information for certain purposes prohibited

Sec. 16. The information contained in the Indiana DNA data base may not be collected or stored to obtain information about human physical traits or predisposition for disease.
As added by P.L.2-2003, SEC.4.

IC 10-13-6-17
Personal information limited

Sec. 17. Personal information stored in the Indiana DNA data base is limited to:
(1) data necessary to:
(A) generate investigative leads; and
(B) support statistical interpretation of test results; and
(2) any other information necessary to allow for the successful implementation of the Indiana DNA data base system.
As added by P.L.2-2003, SEC.4.

IC 10-13-6-18
Expungement of DNA profile

Sec. 18. (a) A person whose DNA profile has been included in the Indiana DNA data base may request expungement of the profile from the DNA data base on the grounds that the conviction on which the authority for inclusion in the Indiana DNA data base was founded has been reversed and the case has been dismissed.
(b) All identifiable information in the Indiana DNA data base pertaining to a person requesting expungement under subsection (a) shall be expunged, and all samples from the person shall be destroyed upon receipt of:
(1) a written request for expungement under subsection (a);
(2) a certified copy of the court order reversing and dismissing the conviction; and
(3) any other information necessary to ascertain the validity of the request.
(c) Upon expungement of a person's DNA profile from the Indiana DNA data base, the superintendent shall request expungement of the person's DNA profile from the national DNA data base.
As added by P.L.2-2003, SEC.4.

IC 10-13-6-19
Access to DNA data base

Sec. 19. (a) Access to the Indiana DNA data base is limited to federal, state, and local law enforcement agencies through their servicing forensic DNA laboratories.
(b) The superintendent shall take appropriate measures to ensure that the

Indiana DNA data base is protected against unauthorized access.
As added by P.L.2-2003, SEC.4.

IC 10-13-6-20
Denial of privileges due to failure to follow quality control and privacy standards

Sec. 20. The superintendent may deny the privilege of a laboratory performing forensic DNA analysis within Indiana to exchange DNA identification records with federal, state, or local criminal justice agencies if required quality control and privacy standards described in this chapter for the Indiana DNA data base are not met by the laboratory.
As added by P.L.2-2003, SEC.4.

IC 10-13-6-21
Unlawful tampering

Sec. 21. A person who knowingly or intentionally without lawful authority tampers with or attempts to tamper with any DNA sample or a container collected under section 10 of this chapter commits a Class D felony.
As added by P.L.2-2003, SEC.4.

IC 10-13-6-22
Unlawful use of data base information or DNA samples

Sec. 22. A person who knowingly or intentionally disseminates, receives, or otherwise uses or attempts to use information in the Indiana DNA data base or DNA samples used in DNA analyses, knowing that such dissemination, receipt, or use is for a purpose other than authorized by law, commits a Class A misdemeanor.
As added by P.L.2-2003, SEC.4.

Appendix 2

ARTICLE 8. INDIANA DNA DATA BASE

Rule 1. Application and Administration

240 IAC 8-1-1 Application of article

Authority: IC 10-11-2-10; IC 10-13-6-8

Affected: IC 10-13-6

Sec. 1. This article governs the administration of the Indiana DNA data base established by IC 10-1-9 *[IC 10-1 was repealed*

by P.L.2-2003, SECTION 102, effective July 1, 2003. See IC 10-13-6.]. (State Police Department; 240 IAC 8-1-1; filed Apr 23, 1998,

9:25 a.m.: 21 IR 3333; readopted filed Oct 6, 2004, 5:10 p.m.: 28 IR 677)

240 IAC 8-1-2 Administration

Authority: IC 10-11-2-10; IC 10-13-6-7; IC 10-13-6-8

Affected: IC 10-13-6

Sec. 2. The commander of the state police laboratory has responsibility for the administration of the Indiana DNA data base

established by IC 10-1-9 *[IC 10-1 was repealed by P.L.2-2003, SECTION 102, effective July 1, 2003. See IC 10-13-6.]* subject to

the authority and approval of the superintendent. *(State Police Department; 240 IAC 8-1-2; filed Apr 23, 1998, 9:25 a.m.: 21 IR*

3333; readopted filed Oct 6, 2004, 5:10 p.m.: 28 IR 677)

Rule 2. Definitions

240 IAC 8-2-1 Applicability

Authority: IC 10-11-2-10; IC 10-13-6-8

Affected: IC 10-13-6

Sec. 1. The definitions in this rule apply throughout this article. *(State Police Department; 240 IAC 8-2-1; filed Apr 23, 1998,*

9:25 a.m.: 21 IR 3333; readopted filed Oct 6, 2004, 5:10 p.m.: 28 IR 677)

240 IAC 8-2-2 "DNA" defined

Authority: IC 10-11-2-10; IC 10-13-6-2; IC 10-13-6-8

Affected: IC 10-13-6

Sec. 2. "DNA" means deoxyribonucleic acid. DNA is located in the nucleated cells and provides an individual's personal

genetic blueprint. DNA encodes genetic information that is the basis of human heredity and forensic identification. *(State Police*

Department; 240 IAC 8-2-2; filed Apr 23, 1998, 9:25 a.m.: 21 IR 3334; readopted filed Oct 6, 2004, 5:10 p.m.: 28 IR 677)

240 IAC 8-2-3 "DNA analysis" defined

Authority: IC 10-11-2-10; IC 10-13-6-3; IC 10-13-6-8

Affected: IC 10-13-6

Sec. 3. "DNA analysis" means an identification process in which the unique genetic code of an individual that is carried by

the individual's DNA is compared with the genetic codes of another individual. *(State Police Department; 240 IAC 8-2-3; filed Apr*

23, 1998, 9:25 a.m.: 21 IR 3334; readopted filed Oct 6, 2004, 5:10 p.m.: 28 IR 677)

240 IAC 8-2-4 "DNA profile" defined

Authority: IC 10-11-2-10; IC 10-13-6-4; IC 10-13-6-8

Affected: IC 10-13-6

Sec. 4. "DNA profile" means the results of all DNA identification tests on an individual's DNA sample. *(State Police*

Department; 240 IAC 8-2-4; filed Apr 23, 1998, 9:25 a.m.: 21 IR 3334; readopted filed Oct 6, 2004, 5:10 p.m.: 28 IR 677)

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240 IAC 8-2-5 "DNA sample" defined

Authority: IC 10-11-2-10; IC 10-13-6-6; IC 10-13-6-8

Affected: IC 10-13-6

Sec. 5. "DNA sample" means a blood, tissue, or other body fluid sample:

(1) provided by a person with respect to offenses covered by IC 10-1-9 *[IC 10-1 was repealed by P.L.2-2003, SECTION 102,*

effective July 1, 2003. See IC 10-13-6.]; or

(2) submitted to the state police laboratory under IC 10-1-9 *[IC 10-1 was repealed by P.L.2-2003, SECTION 102, effective*

July 1, 2003. See IC 10-13-6.] for analysis or storage, or both.

(State Police Department; 240 IAC 8-2-5; filed Apr 23, 1998, 9:25 a.m.: 21 IR 3334; readopted filed Oct 6, 2004, 5:10 p.m.: 28

IR 677)

240 IAC 8-2-6 "Qualifying offender" defined

Authority: IC 10-11-2-10; IC 10-13-6-8; IC 10-13-6-10

Affected: IC 10-13-6; IC 35-42-4-6; IC 35-43-2-1

Sec. 6. "Qualifying offender" means a person convicted of a felony under IC 35-42 (offense against the person), IC 35-43-2-1

(burglary), or IC 35-42-4-6 (child solicitation):

(1) after June 30, 1996, whether or not sentenced to a term of imprisonment; and

(2) before July 1, 1996, if the person is held in jail or prison on or after July 1, 1996.

(State Police Department; 240 IAC 8-2-6; filed Apr 23, 1998, 9:25 a.m.: 21 IR 3334; readopted filed Oct 6, 2004, 5:10 p.m.: 28

IR 677)

240 IAC 8-2-7 "Superintendent" defined

Authority: IC 10-11-2-10; IC 10-13-6-7; IC 10-13-6-8

Affected: IC 10-13-6

Sec. 7. “Superintendent” means the superintendent of the state police department or the superintendent's designee. *(State Police*

Department; 240 IAC 8-2-7; filed Apr 23, 1998, 9:25 a.m.: 21 IR 3334; readopted filed Oct 6, 2004, 5:10 p.m.: 28 IR 677)

Rule 3. Collection and Submission of Samples

240 IAC 8-3-1 Responsibilities

Authority: IC 10-11-2-10; IC 10-13-6-8; IC 10-13-6-10

Affected: IC 10-13-6

Sec. 1. The department of correction shall collect a DNA sample from a qualifying offender, if the qualifying offender has not

previously had a sample collected, and:

(1) is serving a term of incarceration in a facility under the control of the department of correction; or

(2) is transferred to a facility under the control of the department of correction.

(State Police Department; 240 IAC 8-3-1; filed Apr 23, 1998, 9:25 a.m.: 21 IR 3334; readopted filed Oct 6, 2004, 5:10 p.m.: 28

IR 677)

240 IAC 8-3-2 Approved procedure

Authority: IC 10-11-2-10; IC 10-13-6-8; IC 10-13-6-12; IC 10-13-6-17

Affected: IC 10-13-6

Sec. 2. (a) DNA samples shall be collected in a medically approved manner by a physician, registered nurse, licensed

vocational nurse, licensed clinical technologist, or other person at the direction of a physician or under a protocol approved by a

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physician.

(b) Procedures used by the department of correction to extract and obtain liquid blood samples shall be with the approval and

at the direction of the medical director of the department of correction. *(State Police Department; 240 IAC 8-3-2; filed Apr 23, 1998,*

9:25 a.m.: 21 IR 3334; readopted filed Oct 6, 2004, 5:10 p.m.: 28 IR 677)

240 IAC 8-3-3 Collection guidelines

Authority: IC 10-11-2-10; IC 10-13-6-8; IC 10-13-6-10; IC 10-13-6-11

Affected: IC 10-13-6

Sec. 3. (a) The following guidelines apply to the collection of blood samples for submission to the state police laboratory for

the Indiana DNA data base:

(1) Blood samples shall only be drawn from qualifying offenders for use as DNA samples.

(2) A qualifying offender is not required to submit a blood sample if doing so would present a substantial and an unreasonable

risk to the offender.

(3) Blood samples shall be collected using vacutainer tubes containing EDTA preservative or by a finger stick procedure using

stain cards.

(4) After collection of the blood sample, a label approved by the state police laboratory shall be affixed to the blood sample

container; this label shall be completed with required information, including the name and inmate number of the qualifying

offender.

(5) Information shall be recorded on a form approved by the state police laboratory to accompany each blood sample to include

the following:

(A) The qualifying offender's:

(i) name;

(ii) inmate number; and

(iii) inked right thumb print.

(B) If the qualifying offender has no right thumb, then the inked print of another finger and a written notation identifying

the finger used shall be recorded.

(6) The blood sample of the qualifying offender shall be placed in a container approved by the state police laboratory.

(7) An initialed integrity seal shall be placed across the lid of the approved container.

(8) Blood samples may be refrigerated for up to seven (7) days before submission to the state police laboratory.

(9) Blood samples shall be refrigerated until delivery to the state police laboratory.

(b) When it is not possible to obtain a blood sample, the department of correction shall obtain an oral swab.

(c) The following guidelines apply to the collection of oral swabs for submission to the state police laboratory for the Indiana

DNA data base:

(1) Oral swabs shall only be collected from qualifying offenders for use as DNA samples.

(2) Oral swabs shall be collected using sterile cotton tipped applicators.

(3) The following procedure shall be used to collect a sample with an oral swab:

(A) Remove a sterile tipped applicator from its package.

(B) Swab the inner surface of the cheek using a circular motion and complete fifteen (15) to twenty (20) circles.

(C) Repeat the steps in clauses (A) and (B) with a second sterile tipped applicator.

(D) Place the used swabs into two (2) separate envelopes approved by the state police laboratory for the collection of

DNA samples by oral swabs and place the two (2) smaller envelopes into a larger envelope approved by the state police

laboratory.

(4) A label approved by the state police laboratory shall be affixed across the flap of the larger envelope; this label shall be

completed with required information, including the name and inmate number of the qualifying offender.

(5) Information shall be recorded on a form approved by the state police laboratory to accompany each oral swab sample to

include the following:

(A) The qualifying offender's:

(i) name;

(ii) inmate number; and

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(iii) inked right thumb print.

(B) If the qualifying offender has no right thumb, then the inked print of another finger and a written notation identifying

the finger used shall be recorded.

(6) Oral swab samples may be refrigerated for up to seven (7) days before submission to the state police laboratory.

(7) Oral swab samples shall be refrigerated until delivery to the state police laboratory.

(State Police Department; 240 IAC 8-3-3; filed Apr 23, 1998, 9:25 a.m.: 21 IR 3334; readopted filed Oct 6, 2004, 5:10 p.m.: 28

IR 677)

Rule 4. Quality Control

240 IAC 8-4-1 Quality control testing

Authority: IC 10-11-2-10; IC 10-13-6-8; IC 10-13-6-13

Affected: IC 10-13-6

Sec. 1. The state police laboratory shall perform tests on DNA samples for quality control and assurance. *(State Police*

Department; 240 IAC 8-4-1; filed Apr 23, 1998, 9:25 a.m.: 21 IR 3335; readopted filed Oct 6, 2004, 5:10 p.m.: 28 IR 677)

240 IAC 8-4-2 Quality assurance standards

Authority: IC 10-11-2-10; IC 10-13-6-8; IC 10-13-6-14

Affected: IC 10-13-6

Sec. 2. (a) A laboratory conducting forensic DNA analysis in Indiana shall comply with nationally recognized standards for

quality assurance and proficiency testing.

(b) In the event of competing quality assurance standards, the state police laboratory commander shall identify the nationally

recognized standard that a laboratory conducting forensic DNA analysis in Indiana must implement and follow. *(State Police*

Department; 240 IAC 8-4-2; filed Apr 23, 1998, 9:25 a.m.: 21 IR 3335; readopted filed Oct 6, 2004, 5:10 p.m.: 28 IR 677)

240 IAC 8-4-3 Participation

Authority: IC 10-11-2-10; IC 10-13-6-8; IC 10-13-6-20

Affected: IC 10-13-6

Sec. 3. If a laboratory performing forensic DNA analysis in Indiana fails to meet required quality control standards, the

superintendent shall deny the laboratory the right to exchange DNA identification records with federal, state, or local criminal justice

agencies. *(State Police Department; 240 IAC 8-4-3; filed Apr 23, 1998, 9:25 a.m.: 21 IR 3335; readopted filed Oct 6, 2004, 5:10*

p.m.: 28 IR 677)

Rule 5. Authorized Access

240 IAC 8-5-1 Request for access; denial

Authority: IC 10-11-2-10; IC 10-13-6-8; IC 10-13-6-19

Affected: IC 10-13-6

Sec. 1. (a) A forensic DNA laboratory seeking access to the DNA data base must submit a written request to the commander

of the state police laboratory.

(b) The written request required by subsection (a) must state the following:

(1) The identity of the federal, state, or local law enforcement agency requesting access.

(2) The purpose of the testing to be performed.

(c) A request submitted under subsection (a) that does not identify a requesting law enforcement agency or fails to state a

proper purpose for testing under IC 10-1-9 *[IC 10-1 was repealed by P.L.2-2003, SECTION 102, effective July 1, 2003. See IC 10-*

13-6.] shall be denied. *(State Police Department; 240 IAC 8-5-1; filed Apr 23, 1998, 9:25 a.m.: 21 IR 3335; readopted filed Oct*

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6, 2004, 5:10 p.m.: 28 IR 677)

Rule 6. Expungement

240 IAC 8-6-1 Request for expungement

Authority: IC 10-11-2-10; IC 10-13-6-8; IC 10-13-6-18

Affected: IC 10-13-6

Sec. 1. (a) A person whose DNA profile has been included in the Indiana DNA data base and who is eligible for expungement

under IC 10-1-9-20 *[IC 10-1 was repealed by P.L.2-2003, SECTION 102, effective July 1, 2003. See IC 10-13-6-18.]* may submit

a written request for expungement to the superintendent.

(b) All identifiable information pertaining to the person in the DNA data base and all DNA samples related to the request shall

be destroyed upon receipt of the following:

(1) the written request for expungement under subsection (a);

(2) a certified copy of the court order reversing and dismissing the conviction that made the person a qualifying offender; and

(3) identifying information to include the following:

(A) The full name of the person requesting expungement.

(B) The inmate number and name of the person that contributed the DNA sample correlating to the DNA profile to be

expunged.

(C) The inked right thumb print of the person requesting expungement, or if the person has no right thumb, then the

inked print of another finger and information identifying the finger actually used.

*(State Police Department; 240 IAC 8-6-1; filed Apr 23, 1998, 9:25 a.m.: 21 IR 3336; readopted filed Oct 6, 2004, 5:10 p.m.: 28 IR 677)**

APPENDIX 3 DEFINITIONS

1. Allele – the alternative form of a gene.
2. Allelic Ladder – a set of DNA fragments of the commonly known alleles for each locus. By comparing the samples to the allelic ladder, the correct allele designation may be assigned.
3. Amelogenin – the marker for determining the gender of the individual contributor to a DNA profile.
4. Amplification – using the PCR process to create many copies of a specific DNA sequence(s). An increase in the number of copies of a specific DNA fragment.
5. Analyst Discretion - The use of individual judgment, based on an analyst's training and experience to determine the optimum modes of analysis for sample.
6. Artifact – non-allelic product of the amplification process, an anomaly of the detection process, or a by-product of primer synthesis. A data peak that does not represent a true allele.
7. Autosearch – A CODIS program, which automatically searches all DNA profiles in a user specified index against all profiles in one or more other user specified indexes.
8. Candidate Match – A possible match between two or more DNA profiles discovered by CODIS software.
9. Capillary Electrophoresis – a method to separate DNA fragments based on size using electrical current. The DNA sample is placed in thin tube (capillary) containing gel (polymer) and subjected to high voltage current allowing the DNA fragments to migrate through the tube.
10. Reference Material –material obtained from a known source and collected for purposes of comparison to forensic samples.
11. Combined DNA Index System (CODIS) – refers to the DNA database and its software. It is composed of National (NDIS), State (SDIS), and Local (LDIS) components. It contains DNA profiles form offenders, crime scenes, and includes a missing persons database.
12. CODIS Administrator – An employee of the laboratory responsible for administration and security of the laboratory's CODIS.
13. Contamination – the process of making a sample impure or unusable.
14. Convicted Offender (specimen category) – The known sample from a person who has been convicted of a qualifying offense.
15. Convicted Offender Index – consists of DNA records from offenders convicted of qualifying state crimes and juveniles required by relevant jurisdiction to provide DNA samples.

16. Deduced Missing Person – The DNA profile of a reported missing person that has been generated by examining items purported to belong to the missing person such as a toothbrush.
17. Deoxyribonucleic Acid (DNA) - the genetic material present in the nucleus of most cells. Also located in cell mitochondria.
18. DNA Profile – The genetic constitution of an individual at defined locations (also known as loci) in the DNA.
19. DNA Record – A database record that includes the DNA profile as well as data required to manage and operate NDIS.
20. DNA Sequence - a specific order of base pairs.
21. Electropherogram – the visual representation of the DNA fragments contained in each sample; generated by the analysis software of the Capillary Electrophoresis Instrument.
22. Electrophoresis – the method to separate molecules based on their size by placing them in a medium and applying a electrical current. The molecules will travel through the medium at different rates, the smaller molecules traveling through the medium more quickly than the larger ones.
23. Enzyme – a protein which acts as a catalyst, speeding up a specific chemical reaction without being changed or consumed in the process.
24. Forensic Mixture – DNA results originating from a biological sample found at the scene of a crime that contains DNA contributed from more than one source.
25. Forensic Index – consists of DNA records originating from and associated with an evidence sample from a single source from a crime scene.
26. Forensic Unknown – A DNA profile that originates from a single source biological sample found at the scene of a crime.
27. High Stringency Match – requires all alleles to match.
28. Hit – A confirmed match that aids an investigation and one or more of the case(s) involved in the match are unsolved.
29. Internal Lane Standard (ILS) – a set of DNA fragments of known length(s). The ILS is simultaneously injected with all DNA samples during electrophoresis. This allows accurate measurement of the length of each allele in a DNA sample.
30. Match – A match occurs when CODIS links two or more DNA profiles.
31. Missing Person (specimen category) – The known reference sample from an individual that is missing.
32. Missing Persons Index – consists of DNA records from missing persons and deduced missing persons.
33. Moderate Stringency Match – requires all alleles to match, but the target and candidate profiles can contain a different number of alleles.
34. Mixture – A DNA profile containing 3 or more alleles at more than 2 loci.

35. Negative Amplification Control – used to detect DNA contamination of the amplification reagents, consisting of only amplification reagents without the addition of template DNA.
36. Platform – The type of analytical system utilized to generate DNA profiles, such as capillary electrophoresis and real-time gel.
37. Polymerase – an enzyme that initiates the duplication of a DNA molecule.
38. Polymerase Chain Reaction (PCR) – a process for amplifying (copying) the DNA molecule.
39. Positive Amplification Control – an analytical control sample used to determine if the PCR performed properly consisting of amplification reagents and a known DNA sample.
40. Primer – a short nucleotide fragment of known sequence used to locate its complementary sequence on the DNA molecule for the initiation of PCR. Primers target the specific loci to be amplified.
41. Proficiency Testing – a test to evaluate the competency of an analyst in a specific procedure.
42. Qualified DNA Analyst – A DNA analyst who has satisfied and continues to satisfy the experience, education, training, proficiency testing and continuing education requirements of the FBI Director's Quality Assurance Standards issued in accordance with the DNA Identification Act of 1994, as well as successful completion of a qualifying test prior to beginning casework or databasing responsibilities.
43. Reagent Blank Control – an analytical control sample that contains no template DNA and is used to monitor contamination from extraction to final fragment analysis.
44. Relatives of Missing Persons Index – consists of DNA records from the biological relatives of individuals reported missing.
45. Short Tandem Repeat (STR) – small sections of DNA that contain short segments (2, 3, 4 or more base pairs) which repeat several times. The number of repeat units may vary between individuals. STRs are located between specific genes and are considered non-functional.
46. Source ID – A field in the CODIS software used to indicate whether or not the source of a forensic unknown or forensic mixture is known.
47. Stochastic effect – peak imbalance observed in a locus and/or allele drop-out due to random, disproportionate amplification of alleles in low quality/quantity template samples.
48. Stutter – an artifact that occurs as a by-product of the PCR process. It is observed as a minor peak typically observed one repeat unit smaller than a primary STR allele caused by strand slippage during amplification.
49. Technical Review - an evaluation of reports, notes, data, and other documents to ensure there is an appropriate and sufficient basis for the scientific conclusions.
50. Test Kit – A preassembled set of reagents that allow the user to conduct a specific DNA extraction, quantification, or amplication.

51. Unidentified Human (Remains) Index – The DNA profile developed from the recovered deceased (including body parts and tissue) or an individual who is unidentified (e.g., children who can't and others who won't or refuse to identify themselves).
52. Unidentified Person – The DNA profile developed from the recovered deceased (including body parts and tissue) or an individual who is unidentified (such as persons who can't or refuse to identify themselves).
53. Work Product – the material that is generated as a function of analysis, which may include extracts and amplified product, in tubes or plates, and any aliquots thereof.